FACTORS AFFECTING ENERGY ABSORPTION IN POULTRY

by

SUSAN BULLASS

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TO MY PARENTS

DECLARATION

I hereby declare that this thesis describes the results of my own research, carried out during the period October 1979 and September 1982 at the Agricultural Research Council's Poultry Research Centre, Roslin, Midlothian and has been composed by myself.

ABSTRACT

A variety of inert-additives; sand, kaolin, cement kiln dust, sawdust and cellulose, were studied with respect to their potential use as dietary diluents. Each of the diluents was found to have no effect upon the Apparent Metabolisable Energy (AME) value of a conventional broiler diet, but, with the exception of sand, all depressed chick growth at three weeks of age.

Sand, included in conventional diets at levels of up to 60 g/kg, has been shown to have no effect upon the True Metabolisable Energy (TME) values of the diets nor upon the Endogenous Energy Losses (EEL's) of adult cockerels. The presence of dietary sand has also been shown to significantly improve broiler growth at 56 days of age but feed intakes were increased proportionally and there was, consequently, no improvement in feed conversion efficiency. It was concluded that sand does not act by merely reducing the energy density of a diet.

Cellulose has been shown to have no effect upon the TME value of a conventional diet but does depress chick growth. It was concluded that the growth depression is a consequence of reduced feed intakes. It has also been confirmed that cellulose has no energy available for digestion by poultry.

The role of the avian caeca in digestion was studied by feeding conventional and sand- and cellulose-diluted diets. It was concluded that the caeca play no role in digestion under normal circumstances but may play a compensatory role by conserving energy under conditions of stress.

Guar gum was studied for its effects upon chick growth, the metabolisable energy value of a diet, nitrogen retention and fat digestibility. Growth depression was observed at dietary inclusion levels as low as 7.5 g guar gum/kg. It was concluded that the growth depression was a consequence of a reduction in dietary AME, which itself was a result of an overall reduction in absorption of nutrients, the main effect being upon fat digestibility. The TME value of a conventional diet was found to be unaffected by guar gum when measured by the Sibbald bioassay but it was suggested that this was due to guar gum reducing endogenous energy loss.

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CONTENTS

Table of Contents continued

1. INTRODUCTION

LITERATURE REVIEW

Introduction

The price paid for feedstuffs represents the major proportion of the cost of the production of eggs and poultry meat. Consequently anything that can reduce this price will improve the profitability of the poultry industry.

It has long been claimed that poultry utilise low energy-dense feeds more efficiently than high energy-dense feeds (Hill & Dansky, 1954; Mraz et al. 1957). Low energy ingredients are not, however, automatically cheaper than energy dense ones and because larger quantities are required to provide the same total energy they are not always economical to use (Harms et al. 1974; Rowland & Hooge, 1980). However, it is now thought that feeds of low energy-density can be produced effectively by dilution with nutritionally neutral substances; improving efficiency without raising costs.

In recent years the world has suffered frequently from various grain shortages, resulting in elevated prices. These shortfalls have made it necessary to search for alternative feedstuffs and attempts have been made to use "low cost" industrial by-products and/or the sometimes inferior grains that are mainly produced in the Third World as alternatives to conventional raw materials. Guar meal, for example, can be classed as such an industrial by-product but attempts to use it economically in poultry diets have so far failed due to the presence of growth depressing substances.

In 1954, Hill & Dansky reported that maximum growth rate for chicks could be obtained even when dietary energy concentrations were low. This was achieved by marked increases in the birds' feed consumption. However, it was noted that compensation was not complete the total energy intake being progressively reduced as the dietary energy level was decreased.

It has recently been reported that effective feeds with low energy density can be obtained by dilution of diets with inert substances (e.g. kaolin or sand). These diets had no deleterious effects on weight gains or energy utilisation. Indeed, in some instances performance has been improved (Spandorf et al. 1972; Harms, 1974).

Dietary dilution by this technique may prove to be a more economical method of improving energy utilisation in poultry than the use of ingredients of low energy content.

Sand

Builders' sand, commonly called Brick sand (Rowland & Hooge, 1980), has been used for many years as a filler in diets used for mineral studies. Andrews et al. (1972) reported that the use of builders' sand as a diluent in phosphorus bioassays had no influence upon chick performance and that subsequent evaluation of previous data revealed that the addition of sand to the diets had improved dietary energy utilisation by both chicks and hens.

Damron & Harms (1976) and Damron et al. (1976), formulated a diet to contain the same levels of protein and energy as a basal diet with the inclusion of 2.5% sand. Another diet had the same composition as the diet containing sand but with the sand omitted, providing a 97.5% composition (i.e. 97.5% of the basal). Both the sand-diluted diet and the "no-filler" diet resulted in increased feed conversion efficiency measured in terms of egg-production. The efficiency of energy utilisation was also increased by the addition of 2.5% sand.

Hooge & Rowland (1978) reported that the addition of 5, 10 or 15% sand to a typical diet for laying hens decreased the actual feed consumption (i.e. feed minus diluent) and improved energy utilisation over the 336 day experimental period. Egg production on a hen-day basis was also increased. In a concurrent trial with broiler chicks the addition of 6% sand to a range of diets covering three levels of protein combined with three levels of metabolisable energy (ME), improved adjusted feed conversion (i.e. feed minus sand) and energy utilisation by 2.6%. Although growth was slightly poorer for the birds fed diets containing diluent the addition of sand proved economical because less basal feed was required for a bird to attain a given weight.

It has been suggested that older birds are less likely to be affected adversely by dietary dilution by sand than younger ones, due to their greater body protein and energy stores (Voitle et al. 1974). The addition of 5% sand to the diet of 29-week old broiler breeder pullets reduced egg production but there was no effect upon body weight, egg production, egg size, hatchability or fertility of 66-week old hens. Hogsette et al. (1976) reported that the addition of 5% sand to the diet of broiler breeders had no significant effect upon

egg production or the amount of food consumed per egg. The amount of energy required per egg was however reduced, suggesting a more efficient utilisation of energy by birds fed diets containing sand. An increase in energy utilisation, in terms of egg production, was also indicated in studies by Hooge et al. (1977). Furthermore, the addition of up to 15% sand to the diet of laying hens had no adverse effects on egg production nor on egg weight. Harms & Damron (1973) reported that the addition of 5% sand to the diet of laying hens improved energy utilisation by about 7%, whilst with growing chicks the addition of 5 and 10% sand improved it by 6.55% and 4.74% respectively.

Improved dietary energy utilisation as a result of the addition of sand has also been indicated in experiments with turkey poults (Harms & Voitle, 1977). Dilution of a diet based on maize and soya bean meals by 2.5, 5 and 10% sand had no significant effect upon body weight gains measured at 10, 13 and 21 days of age. Actual feed (i.e. less diluent) efficiency was significantly improved by the addition of 2.5% sand. The efficiency of energy utilisation was significantly increased by each addition but the greatest improvement was shown by birds fed the diet containing 2.5% sand.

Miles et al. (1978,1981) reported that sand could be included in the diets of turkey poults at 2.5% with no adverse effects on either weight gain or feed efficiency. The True Metabolisable Energy (TME) of the substituted diet was higher than that of the basal diet but this difference was not statistically significant.

Sellers et al.(1979,1980) studied the effect of various inert fillers upon broiler performance and reported that the addition of sand at either 2.5 or 5% resulted in no significant changes in body weight gains, feed efficiency or feed consumption. Rowland & Hooge (1980) concluded that at least as much as 6% sand could be added to a broiler starter diet to improve feed utilisation and at the same time have no adverse effects upon body weights. In three experiments the addition of 6% sand to the bird's diet actually improved body weights although the increases were not statistically significant.

The physical properties of sand may be important in determining the effect of its addition to poultry diets. Harms (1974), suggested that the presence of sand in the diet causes greater distribution of

food within the digestive tract, thereby allowing the digestive juices greater access to the feed and support for this hypothesis has been reported by Oluyemi & Harms (1977). In experiments with turkey poults the extent of the improvement of both feed and energy utilisation tended to be greater the smaller the particle size of the sand. Oluyemi et al. (1978) observed that neither sand nor grit accumulated in the gizzard. When finely pulverised it appeared that their use in grinding was limited. Because sand passed through the gizzard more rapidly than grit it was concluded that it was less efficient than grit for grinding. This might be offset by sand passing more quickly into the gut, mixing with the food and allowing greater exposure of food particles to the digestive juices.

It has been postulated (Hooge, 1979) that the addition of sand to poultry diets causes a decrease in heat production, due to dietary induced thermogenesis per unit time. This would result in lower energy loss to the environment in the form of heat, and allow the more efficient utilisation of dietary energy.

Hooge (1979) recommended that sand should be added only at the expense of complete feed. For broilers 5 or 6% sand appears optimum for use in a starter diet containing 23% protein and 13.40 MJ.ME/kg. Hooge further reported that at lower protein and energy levels sand improved feed and energy utilisation but equivalent weight gains were attained only after a slightly longer time than normal.

Overall, results suggest that sand can be successfully added to poultry diets with no adverse effects and may be a valuable way of improving feed utilisation. Feed and, therefore, cost of production are reduced.

Kaolin

Various reports have attributed seemingly nutritional qualities to members of the clay family. Thus improvements in rate of weight gain, feed efficiency, egg shell quality, egg production, carcass quality and animal health have been demonstrated at one time or another. Inclusion levels have generally ranged from 1 to 10%, either as a diluent to the complete diet or replacing some ingredient, generally part of the grain fraction. Animals studied have included poultry and livestock such as hogs, sheep, beef and dairy cattle

(Jordan 1953, 1954; Erwin et al. 1957). Results have rarely been spectacular, showing either no significant difference or a smaller depression than that expected due to the actual dilution.

There are many clays and clay-like minerals in nature, generally referred to as kaolins, bentonites, vermiculites, silicates or, in the case of mixed bodies, as "clay". Physically most are sheet-like, complex silicates with variable contents of exchangeable cations including Na^+, Ca^{++}, Mg^{++} and K^+. Their behaviour in water varies - some swell, others gelate - but generally the crystal plates separate to some degree, resulting in a very small particle size (much less than 1μ) with an enormous surface area (Ousterhout, 1970). This may explain the increase in feed utilisation observed by mechanisms similar to those previously suggested for sand. Various workers have suggested that the use of clays in poultry diets may have improved feed utilisation by slowing feed passage time through the intestine thereby allowing more complete digestion and greater nutrient absorption (Quisenberry & Bradley, 1964; Almquist et al. 1967; Kurnick & Reid, 1960; Ousterhout, 1967).

Ousterhout (1970) suggested that, in ruminants at least, the action of kaolin depends upon its ability to stabilise gut mobility during stress. Thus the excessively rapid passage of ingesta, which might be expected to limit absorption and/or the action of gut micro-flora, is prevented. He pointed out that such action would have little or no effect on feed efficiency during normal health but could have considerable effect under conditions of stress.

Ousterhout (1967) included a variety of kaolins in broiler diets at inclusion levels ranging from 0.5 to 40% and reported that up to 16% resulted in improved energy utilisation. In a separate experiment battery broiler chicks received diets containing between 2.5 and 8% kaolin either as a complete dietary diluent or as a substitute for maize. Energy utilisation was improved to the extent of about half the percentage inclusion of kaolin. Ousterhout concluded that the kaolin acted by slowing the rate of passage of ingesta through the gut leading to improved digestion and absorption. In these trials the dietary kaolin had no effect on either the weight gained nor the body composition.

Matterson et al. (1972) included kaolin at 6% in a standard

broiler diet to White Plymouth Rock cockerels from the age of 1 day to 4 weeks. There were no significant differences in weight gains but feed efficiency was significantly improved, suggesting an action other than dilution. In two feed trials, performance on a regular broiler diet was compared to that from one diluted by 5% kaolin. Weight gains were unaffected but the gain/feed ratio was improved by about 6%.

Spandorf et al. (1972) reported that the inclusion of 2.5 or 5% kaolin increased feed conversion by 7.9 and 7.4% respectively, compared to the basal diet. Feed efficiency in terms of egg production was also improved. In a separate trial pullets were reared on diets containing either 0 or 6% from 8 to 20 weeks and then fed on diets containing 0, 2.5 or 5% kaolin for a further 20 weeks. Regardless of rearing diet egg production was better from birds fed diets containing kaolin. Furthermore, birds reared on the kaolin-substituted diets performed best. Feed efficiency, however, was unaffected.

Work involving kaolins has shown relatively inconsistent results, but generally the inclusion of kaolin in the diet leads to improved feed conversion efficiency.

Cement Kiln Precipitator Dust

A growth promoting action has recently been ascribed to cement kiln precipitator dust, based upon observations made with ruminants and monogastric animals including chicks. Precipitator dust is a complex, calcium-rich mixture of minerals that results when hot air is pulled out of the cement kiln (Table 1.1). It has none of the alkalis and hardeners that are responsible for cement setting (Wheeler, 1978).

Early work of Wheeler & Oltjen (1978) was stimulated by undocumented reports from Georgia farmers of improved weight gains by growing steers raised on a diet of soya bean meal, straw and maize when cement kiln dust (CKD) was added as a source of calcium. A diet containing 3.5% CKD was fed to 7 steers with 7 control animals simultaneously fed a diet devoid of CKD. After 112 days the test steers had gained 28% more weight than the controls whilst consuming 21% less feed. The extra weight was shown to be all meat which was of a higher quality than that from the controls.

Table 1.1. Analysis of Cement Kiln Dust from Georgia (Wheeler, 1978).

Item	%	Item	ppm.
Calcium	27.31	Copper	42
Phosphorus	0.40	Cobalt	3
Sodium	0.23	Zinc	145
Potassium	0.40	Manganese	152
Magnesium	0.52	Selenium	17
Iron	1.11	Molybdenum	5
Sulphur	2.33	Chromium	110
Chlorine	1.10	Lithium	64
Aluminium	4.16	Strontium	15
		Mercury	15
		Cadmium	4
		Arsenic	7
		Lead	124

Similar results were observed with a second group of 32 steers and also with 60 lambs (Roginski & Wheeler, 1978). Laboratory rats were also reported gaining 23% more weight when fed a semi-purified diet containing 1% CKD. Numerical increases in weight gains were recorded after feeding diets with levels of 2 and 7% CKD.

Results compatible with the findings of Wheeler & Oltjen have been observed with chicks (Kienholz, 1978). Four different Portland cement kiln dusts were included at 1.5% in the diet of Indian River male broiler chicks from 8 to 29 days of age. Diets containing three of these dusts resulted in significant increases in body weight gain and feed efficiency. It appears therefore, that the growth promoting action of cement kiln dust is not restricted to ruminants.

Veltmann & Jensen (1979) could attribute no improvement in growth or feed efficiency to CKD. A practical broiler starter ration was diluted with CKD up to 9% but no significant improvements in performance were found, and, at the higher levels, severe outbreaks of rickets occurred. When 3% or more CKD was added to a broiler starter diet that was unbalanced in calcium and phosphorus, growth depression and a rachitic condition resulted (Veltmann & Jensen 1980). With diets of equal calcium and phosphorus contents the addition of CKD still did not result in an improved performance of chicks and the authors concluded that CKD does not contain factors that will enhance the performance of poultry fed practical diets.

There have been no immediate explanations for the reported growth response to cement kiln dust. Wheeler & Noller (1976) demonstrated the beneficial effect of limestone buffer upon the digestibility of ruminant diets but Wheeler & Oltjen (1978) suggested that this action could account for no more than 30 or 40% of the observed growth response by steers to CKD. Another suggestion is that the high temperatures to which the kiln dust is exposed (1,500°C) could influence mineral availability. It is also possible that improved digestion may result from the extreme fineness of kiln dust (<6μ) and its correspondingly large surface area. The particles may exert a physical action within the gastro-intestinal tract, possibly by breaking up food particles and allowing their greater access to digestive enzymes (or rumen micro-organisms).

The prospect for cement kiln dust as an ingredient in livestock feed remains uncertain. A possible area for concern is the toxic nature of some of its components (Table 1.1). Although the elevated intake of lead, arsenic, mercury, cadmium and selenium may not result in an apparent toxicity, the possible accumulation of these elements in vital organs or their transfer to the egg and embryo is potentially dangerous. There is also a considerable variation in the level of toxic minerals among kiln dusts from different sources. For example, lead can vary in quantity from 124 ppm to 4000 ppm (Wheeler & Oltjen 1978). Such uncertainty must be taken into account when evaluating kiln dust as a possible feed ingredient for livestock.

Fibre

The present day success of the broiler industry is due in no small way to the earlier development of high-efficiency feeds. High-efficiency feeds are also known as "high energy - low fibre" feeds and are produced with the reduction or elimination of feed ingredients containing an appreciable amount of crude fibre. The reduction of the crude fibre content of a diet generally means that the digestibility of the diet is increased. In recent years however, there has been some evidence that poultry can tolerate higher levels of fibre in their diets (Hill & Dansky, 1954).

The value of dietary fibre for ruminants is well documented but little is known of its importance to the nutrition of mono-gastric animals such as the chicken. Early studies have indicated that microbial fermentation of dietary fibre in the caeca contributes only little, if any, available energy to non-ruminants.

There is still considerable controversy as to what compounds contribute to the dietary fibre content of feedstuffs and there is even considerable debate as to the nomenclature that should be used to describe this fraction (Van Soest & Robertson, 1976). The fibrous portion of feedstuffs is generally considered to comprise cellulose, lignin and hemicellulose, although other indigestible polysaccharides are also sometimes included (Scott et al. 1976). Such compounds are generally considered to be entirely indigestible by poultry and other non-ruminants. This concept of "total indigestibility" however can be questioned.

Halnan (1930) found that the digestibility of the indigenous fibre in various feedstuffs, including oats, was rarely above 10% and concluded that the digestibility of the fibre in feedstuffs could not be considered to make a contribution to the nutritive value of the feedstuff. On measuring the percentage digestibility of the organic matter and nitrogen-free extract and comparing these with the fibre content of the feedstuff, Halnan found that their digestibilities were inversely proportional to the fibre content. In the same experiment he found no evidence of an adverse effect of fibre on the protein and fat digestibilities. Halnan concluded that fibre served only to regulate the volume of diets and therefore the amount of nutrients consumed. Mangold (1934) however, reported that 20 to 30% of the fibre of grain could be digested by poultry. Definition differences as to what constitutes fibre offers the likeliest explanation for this apparent contradiction.

It has generally been thought that excessive amounts of fibre in poultry diets reduce feed efficiency, growth and egg production. Heuser et al. (1945) concluded that low-fibre diets, containing about 2.65% fibre, were more efficient than high-fibre diets (5.35% fibre) due to their greater nutrient availability. Scott et al. (1947) replaced 25, 50, 75 and 100% maize with pulverised oats and reported a progressive depression in growth and efficiency of feed utilisation as the quantities of fibre increased. Vlcck (1970), by the addition of 2 to 8% ground barley straw, increased the fibre content of typical starting and growing diets from 5.5 and 6.5% to 7.9 and 8.8%. After periods of 5 and 9 weeks, male chicks fed the 7 and 8% straw diets were significantly lighter than other groups; feed efficiency also decreased as the dietary fibre increased. El-Kotoury et al. (1973) found that egg weight was unaffected by increasing fibre levels but feed conversion efficiency, in terms of egg numbers, was reduced by the inclusion of 7% dietary fibre in trials with Baladi White hens and by 12.5% fibre with Rhode Island Reds.

Conversely, it has also been reported that fibre may have no adverse effects when incorporated into poultry diets. Morris et al. (1932) reported that fibre at up to 9% inclusion had no harmful effects on chick mortality, growth rate, feed consumption, age of

maturity or egg production. Kubota et al. (1967) reported a significant decrease in food intake and egg production when the birds' diet contained 7% crude fibre but there was no difference in energy utilisation among diets containing 2, 4.5 and 7% fibre.

The physical characteristics of fibre are claimed to enable it to modify gut function. Thus its water-holding capacity increases gut transit time and its absorptive properties may alter the availability of some nutrients (Eastwood, 1973). It can also have an abrasive action on the intestinal wall (Hallsworth & Coates, 1962) although the significance of this observation has been questioned (Sibbald, 1981). Hedge et al. (1977) reared chicks on a low residue diet with and without fibre supplements in the form of wheat bran, wheat straw and bagasse. In each of seven experiments 10% wheat bran improved growth and in three experiments feed conversion efficiency was also increased. It was found, however, that the growth promoting effect of the bran was not directly due to its fibre content because the same results were not obtained when a diet containing an equivalent amount of fibre provided by wheat straw was fed. The authors suggested that wheat bran may contain a growth-promoting agent, as yet unidentified.

The bulk of intestinal contents is increased by the water-holding capacity of fibre. If this resulted in an increase in gut dimensions absorption could well be improved. Conversely, an abrasive action might be expected to impair absorption. Hedge et al. (1977) however, found no conclusive evidence supporting either hypothesis in their experiments.

Cellulose is probably the most common form of fibre used in laboratory experiments involving animals that do not produce the enzyme cellulase. It is the earths' most abundant organic compound, forming the fibrous tissue of the vast majority of plant materials. Chemically, cellulose is a linear polymer of glucose connected by β(1-4)-glycosidic linkages (Lang & Briggs 1976). Within the context of dietary fibre the major properties of cellulose are:- (1) its susceptibility to hydrolysis by enzymolysis and (2) its capacity to absorb water (Southgate, 1976). Cellulases are widely distributed amongst fungi and bacteria and may be involved in the germination of the seeds of higher plants. However, no cellulase

activity has been demonstrated in digestible secretions of either mammals or poultry. The breakdown of cellulose such as occurs in ruminants is attributed to the gut microflora which secrete cellulase. Many mammals that harbour cellulolytic bacteria have evolved specialised anatomy, such as rumens, elongated intestinal tracts and enlarged caeca. All of these features permit prolonged contact between the enzyme and the substrate (Lang & Briggs, 1976).

Briggs & Davis (1947) found that the addition of a purified source of cellulose to a synthetic diet improved the growth rate of chicks and was "otherwise beneficial". Retarded growth was only observed among birds fed diets with quantities of cellulose greater than 20%. At four weeks, average weights and feed efficiencies were significantly increased by those diets containing 5, 10 and 15% cellulose, implying that the cellulose was not nutritionally neutral and functioning simply as bulk. The authors proposed either that the hydrolysis of cellulose in the gastro-intestinal tract was producing growth promoting substances or that the action of cellulose is physical and, in some way, was aiding the absorption of nutrients.

Peterson et al. (1954) fed cellulose at 0, 12, 24, 36 and 48% in diets containing four levels of protein to White Leghorn chicks from 9 to 29 days of age. Food consumption (and, as a result, growth rate) was increased by replacing glucose with moderate amounts of cellulose. At very high cellulose levels the energy content was so low that the birds could not consume sufficient feed to meet their energy requirements and growth was depressed. It was suggested that at the lower levels of inclusion growth rate was determined by protein intake. Growth could be increased either by raising the protein level of the diet or by decreasing the density of the calculated ME (i.e. by increasing the bulk of the diet) which, by increasing the food consumption, resulted in a higher protein intake and increased growth.

Purified cellulose can be added to a diet without changing the balance of nutrients. Because cellulose cannot be digested by the chick (Tasaki & Kibe, 1959) it is assumed to serve only as a diluent. Tasaki & Kibe reported that the total amount of cellulose ingested by chickens can be recovered in the faeces, though not

necessarily in its original form; α-cellulose was found to be partially degraded to β-cellulose and excreted as such, whereas β- and γ-cellulose were excreted unchanged. The authors pointed out that these results conflict with another report (Saito et al. 1959) that the growth promoting action of cellulose was due to energy production from cellulose digestion. Saito et al. reported significantly higher weight gains when diets containing 3.5, 9.5, 16.5 and 26.5% cellulose were fed to White Leghorn chicks, compared to those fed the control diet containing no cellulose; there were no significant differences among the cellulose treatments.

Dvorak & Bray (1978) reported that the addition of between 10 and 45% cellulose to a chick diet caused a linear increase in feed intake and a concomitant depression in growth. Furthermore the non-digestible material reduced the utilisation of the basal portion of the diet.

A marked reduction in growth rate was reported by Bayer et al. (1978) after the addition of 6% cellulose to a chick starter diet at the expense of maize. From three to six weeks of age the birds fed the control diet had a significantly higher feed efficiency than those on the cellulose-diluted diet. The observed increase in water intake and the lower dry matter content of the crops suggested a faster gut transit time. This could account for the extent of the reduction in growth which is otherwise not fully explained by the 6% dilution by cellulose.

With the use of a nutrient balance technique and isoenergetic diets, Begin (1961) demonstrated that the addition of wood pulp cellulose had neither a depressing nor stimulating effect upon chick growth. Digestibility studies showed that cellulose was not utilised by the chicks and had no apparent influence upon nitrogen retention.

Yoshida & Hoshii (1970) reported that cellulose products had neither biologically available energy nor growth-promoting or -depressing effects and suggested that the neutral properties of cellulose allow its safe use as a diluent in poultry feeds, in nutritional experiments and in energy bioassays.

Sawdust, in the form of spruce wood flour, was reported to have no deleterious effects on chicks when included in the diet at 20.2% (Ewing 1947). Indeed, there was a slight increase in growth at the

age of 28 weeks. Davis & Briggs (1948) substituted glucose in a purified diet by 10, 15, 20 and 30% of mixed, white and yellow pine, sawdust. Up to 20% was tolerated without any adverse effects. At 30% inclusion the chicks suffered growth retardation and reduced feed utilisation was noted.

El-Abbady et al. (1973) reported increased body weights with low levels of sawdust. The addition of 0, 4.0, 10.5 and 20% sawdust resulted in diets having crude fibre contents of 4.24, 7.12, 11.96 and 16.84%. The 20% inclusion significantly decreased body weight gains in male Baladi Whites at 20 weeks of age.

Similar results were reported by Omar et al. (1973) in a trial involving White Austrian Turkeys. The turkeys were maintained from 4 to 24 weeks on isoenergetic, isonitrogenous diets with sawdust included at 0, 6, 12 and 18%. These diets had crude fibre contents of 4.34, 8.87, 12.55 and 16.99% respectively. The inclusion of 6% sawdust in the diet maintained male body weights but higher inclusion levels depressed growth.

It has been reported that compared to floor-reared broilers cage-reared broilers have smaller gizzards and a less pronounced differentiation between the gizzard and proventriculus. Addition of ground pine shavings to the diet of cage-reared broilers has been found to correct the gizzard-proventriculus problem (Deaton et al. 1976). Up to 6% pine shavings in a general layer diet improved gizzard weights. There were, however, no apparent effects upon body weight or egg production although feed efficiency was increased.

It appears therefore, that low levels of sawdust can be added to poultry diets without detrimental effects and, indeed, may improve feed utilisation.

The absence of cellulase activity in the digestive juices of poultry (Lang & Briggs, 1976) leads to the assumption that any digestion of fibre would be due to the presence of cellulolytic bacteria in the digestive tract. It is likely that any such digestion could only occur in the crop or the caeca where food, or food residues, remain for an appreciable length of time (Halnan, 1930). Halnan (1930) quotes Lippincot as stating that the crop's contents are too acid for such bacterial action which suggests that the caeca provide the most likely site for any possible digestion of fibre in poultry.

The role of caeca in fibre digestibility

The domestic fowl possesses a pair of caeca, blind tubular sacs, positioned at the terminal portion of the ileum to which they are linked by mesentery. In the mature bird each caecum is about 15 to 18 cm in length and consists of a narrow constricted open end with a dilated, thinner walled, blind portion (McNab 1973). The histological structure is similar to that of the small intestine but there is a tendency for a greater abundance of lymphoid tissue.

Very little is known about the role of avian caeca, especially with respect to digestion. The caeca have generally been considered to be of little nutritional importance in the domestic fowl since their removal has no apparent effect on growth rate nor performance.

It has, however, been suggested that a significant amount (Thornburn & Willcox 1965a), if not the majority (Mangold 1934) of the crude fibre eaten by chickens is digested in the caeca. Halnan (1949) quoted reports (Radeff, 1928; Henning, 1929) of caecectomy resulting in much lower digestibility coefficients for the crude fibre of wheat and maize, indicating that at least some digestion of crude fibre takes place in the caeca.

Thornburn & Willcox (1965a) reported that caecectomy reduced the overall digestibility of food dry matter as well as the digestibility of crude fibre. The latter effect, however, is dependent not only on the nature of the food being eaten but also on its crude fibre content. Cellulose digestibility was reduced in individual birds after caecectomy but this effect was not always apparent when compared with intact birds. Digestion of bran and hays in the caecum was demonstrated *in vivo* by experiments involving the use of caecal fistula (Thornburn & Willcox 1965b) but chemical analysis of the bran and hay gave no evidence of the type(s) of compound that was digested. Incubation of caecal material *in vitro* with a polysaccharide substrate (maize starch and xylan) led to the substrates' breakdown into simple soluble carbohydrates and then into acidic metabolites, including pyruvic and lactic acids. The authors suggested that the first stage occurs outside the bacteria which are, nevertheless, responsible for it. The second stage then takes place within the bacteria after absorption of the metabolites of the first stage.

In contrast, Nakahiro et al. (1974) concluded that the caecum does not play an important role in the digestion of crude fibre in chickens. Caecal ligation did not affect the digestibility of crude fibre, cellulose or pentosan when a semi-purified diet containing 12.5% fibrous screenings of Italian ryegrass was fed to chicks. These findings were confirmed in a later experiment involving the use of maize, wheat, copra meal or grass fibre based diets (Nakahiro & Isshiki, 1975).

It has long been recognised that caecal microflora have fermentative abilities. Shrimpton (1963) demonstrated the occurrence of volatile fatty acids (VFA's) and Shrimpton & Stevens (1965) reported that strains of Bacteroides sp. in the caeca ferment glucose with the production of acetic and propionic acid. Annison et al. (1968) confirmed this and also demonstrated the absorption of VFA's into the portal system. VFA's were demonstrated as being the major end-product of fermentation in the digestive tract, the caeca being the main site of production. Caecectomy effectively reduced the level of VFA's in the excreta but did not completely eliminate them and the authors suggested that other regions of the digestive tract may have assumed this role of the caeca. The type and extent of fermentation is thought to depend on the microbial population and the nature and quantity of substrate entering the caeca. Annison et al. (1968) demonstrated the presence of VFA's in portal blood and suggested that absorption of these fermentation products had occurred. The presence of acetate in the peripheral blood indicated to the authors that either the production of acetate in the digestive tract was in excess of the liver's ability to metabolise it or that there was a significant endogenous contribution to the level of circulating acetate. The suggestion was made that the extent of caecal VFA production may be modified under different dietary regimes and, if so, then exogenously derived acetate may become a more significant energy source.

There is evidence that the nature of the diet can alter the size of the caeca and hence affect their function. Lewin (1963) reported that the caecal length of Californian Quail increased in winter to 12.5 cm compared to its summer length of 8 cm. Villi lengths also increased and it was suggested that these changes enable the improved extraction of nutrients from the lower quality, bulky

diet on which the birds depend during winter. The lengths of the small and large intestine and caeca vary seasonally in the Spruce Grouse (Pendergast & Boag, 1973) being longer in the winter when the diet consists mainly of conifer needles than in summer, when fruit is an important item of the diet.

Savory & Gentle (1976a) reported that gut dimensions, including caecal length, of Japanese Quail were significantly increased when the diet contained 20% sawdust but suggested that this was due to the increased feed intake rather than fibre per se, because previously it had been shown that gut structure was unaffected. The results were confirmed in a later experiment (Savory & Gentle, 1976b). When diets were interchanged (i.e. high fibre to low; low fibre to high) feed intakes were adjusted appropriately. Gut dimensions also adjusted, changing at similar rates and reaching the appropriate sizes for the respective diets in 3 to 4 weeks, although feed intakes were adjusted in 8 to 10 days. The authors suggested that the prolonged adjusting period of gut dimensions was to attain the minimum gut size necessary to maintain a desired rate of digestion. It was postulated that, initially, after the change from a low to a high fibre diet the gut would have been too small to accommodate the increased food intake. Digestibility was not however affected, suggesting that the rate of digestion was increased and this would probably have been the stimulant producing the gradual growth of the gut.

It has been concluded (McNab 1973) that caeca are unnecessary for the optimal growth of domestic birds, although they may serve an important role in the wild state enabling the bird to conserve water, nitrogen and energy, and it is possible that they play a compensatory role when the normal digestive processes in the small intestine are impaired.

Guar, Cyamopsis tetragonoloba, is a drought resistant legume which has been extensively grown for many centuries in the Indian subcontinent to provide livestock and human feed. Although India is still the largest producer of guar it has also been successfully introduced to many other areas of the world, including North America. A major interest in guar developed during World War II when it was realised that guar gum was a suitable substitute for carob seed gum which was previously used by industry and in short supply because of the war. This stimulated the growth of guar gum extraction plants. Gum is extracted from guar seeds by a dry milling process and the by-product is termed guar meal (Verma, 1977).

Guar meal can contain approximately 47% crude protein which is rich in lysine and the sulphur containing amino acids. It should, therefore, be a source of high quality protein. However, its inclusion in poultry diets has been severely restricted due to problems of palatability and certain, so far unidentified, nutritional limitations. Various feeding experiments with guar meal have demonstrated low feed consumption, inhibition of growth, high mortality rates and the occurrence of sticky droppings. The poor performance of chicks fed on guar meal has been largely attributed to the presence of residual guar gum (Kratzer & Vohra, 1963; Nagpal, 1968; Verma, 1977).

Guar gum is a polysaccharide consisting of a backbone of the pyranose units of D-mannose (64%) with D-galactose (36%) sidechains. The most likely structure appears to be one in which all D-mannose units in the chain are connected by β 1-4-glycosidic linkages with α 1-6 linked D-galactose side chains (Rafique & Smith, 1950; Whistler & Smart, 1953). Fig.1.1 is the Whistler & Smart representation of the guar gum unit structure.

It has been reported that guar meal contains two major deleterious components: (1) residual gum, and (2) anti-trypsin factor (Bakshi et al. 1964) but the presence of a haemagglutinin and saponin has also been suggested (Verma, 1977). It is, however, generally considered that the residual gum is the major cause of the inhibition (Vohra & Kratzer, 1964; Anderson & Warnick, 1964).

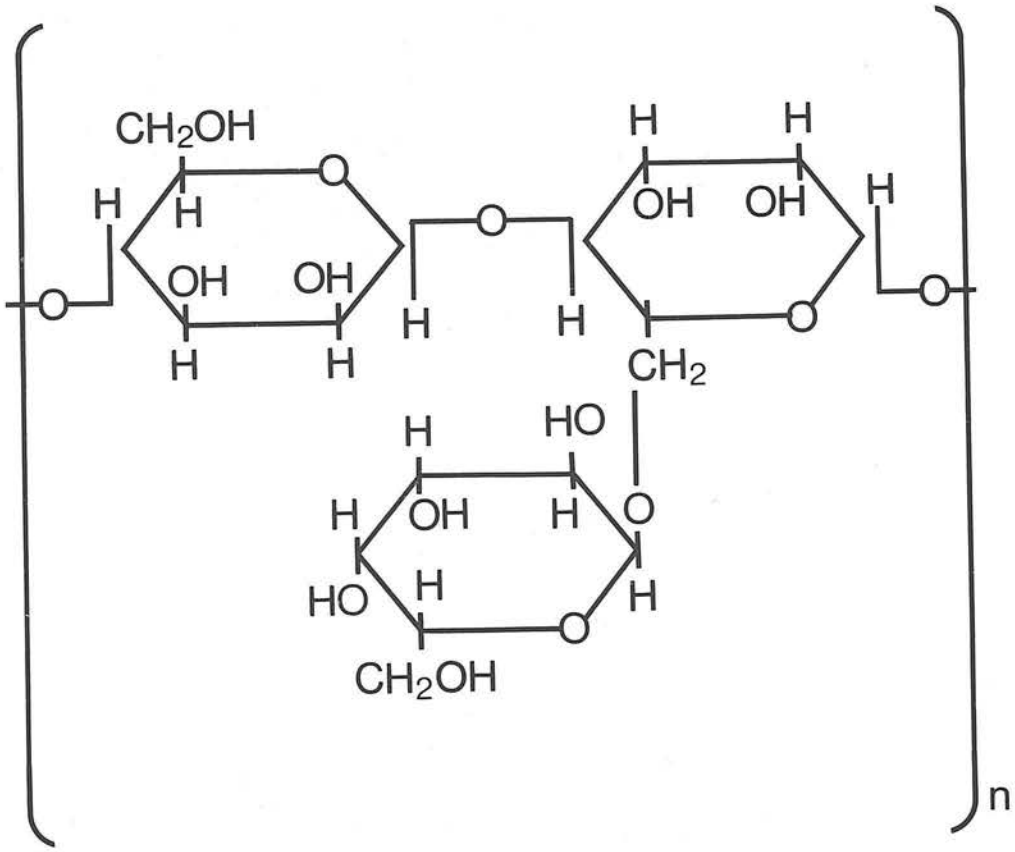

Fig. 1.1 Whistler & Smart (1953) representation of the guar gum unit structure.

Values reported for residual gum in guar meal vary considerably. Nagpal et al. (1971) found gum residues of 18%, Anderson & Warnick (1964) reported 14% while Verma (1977) found gum at only 4 to 6%. Sathe & Bose (1962) found that 12.5% guar meal in growing chick rations depressed growth rate and feed efficiency and Vohra & Kratzer (1964) reported a significant depression in growth with only 7.5% guar meal. If it is assumed that 14% of guar meal is gum, this suggests that gum has deleterious properties certainly at the 2% inclusion level and perhaps even at as low as 1%.

Vohra & Kratzer (1964) added 2% guar gum to the normal stock diet of growing chicks and found that it depressed their growth to 61-67% of the control birds. The addition of 2% cellulose did not have this effect and so the authors suggest that the result was not due simply to dilution of the diet. Decreasing the level of gum proportionately reduced the extent of growth inhibition but some depression of growth could still be detected when only 0.25% gum was present in the diet.

It has been reported that 2% guar gum added to a basal diet at the expense of sorghum depressed growth and significantly reduced nitrogen retention, fat absorption and the metabolisable energy (ME) of the whole diet when fed to growing chicks (Kratzer et al. 1967). When a high-protein diet was used the growth depression was even greater, an observation which led the authors to suggest that the major effect was upon nitrogen utilisation. They also suggested that the significant reduction in ME was due to a combination of the effect on nitrogen retention and fat absorption.

White et al. (1978) showed a linear reduction in weight gains accompanied by an increase in the occurrence of sticky faeces when 1.5% guar gum was added to both maize and barley based diets. Vohra et al. (1979) found that guar gum, substituted for maize starch at a level of 2%, significantly inhibited the growth of chickens and Japanese quail. The digestibility of the diet was significantly lowered in both species, with quail also showing reduced feed intake.

Various attempts have been made to overcome the deleterious effects of guar meal including enzyme supplementation (Anderson & Warnick, 1964; Vohra & Kratzer, 1965)and physical treatment such as autoclaving (Borcher & Ackerson, 1950; Vohra & Kratzer, 1964a).

Anderson & Warnick (1964) found that growth rate of chicks fed a diet containing 10% guar meal was significantly improved by the addition of small amounts (1% or less) of the following enzyme supplements:- Rhozyme T22; Rhozyme CL; Lipase B and Cellulase 36. Lipase B, Rhozyme CL and Cellulase 36 also significantly improved the feed efficiency and reduced the incidence of sticky droppings. (Rhozyme enzymes exhibit hemicellulase activity).

Vohra & Kratzer (1965) tested the effect of various enzymes when added to diets containing 20% toasted or autoclaved guar meal. Keratinase, Diazyme, Pectinase and fungal Amylase were not effective but levels of 0.1 to 0.2% of cellulases, which break down hemicellulose and gums, caused a marked improvement in growth.

Borcher & Ackerson (1950) and Vohra & Kratzer (1964) both reported that raw guar meal could be slightly improved by autoclaving, but without completely overcoming its inhibitory properties. Vohra & Kratzer also reacted the autoclaved meal with an enzyme isolated from sprouted guar and found that this overcame the growth inhibition produced by untreated guar meal.

Bakshi et al. (1964) heated guar meal at 120°C for one hour, followed by steam injection for 15 minutes and found that the processed meal could replace soya bean meal to the extent of 10% without any of the previously noted adverse effects. Patel et al. (1980) demonstrated that both γ-irradiation and pectic enzyme supplementation of guar gum improved growth and almost eliminated the growth depression caused by diets containing 2% guar gum. A combination of γ-irradiation and pectic enzyme supplementation also significantly reduced faecal stickiness.

Various explanationshave been suggested for the mechanism of the action of guar gum. The poor performance of chickens subjected to guar meal may be due to reduced absorption of nutrients through the intestinal wall because of the formation of a protective layer of gum on its surface (Katoch et al. 1971). In humans guar gum has been found to reduce post-prandial glycaemia. Suggested reasons for this vary but it is generally accepted that there is a reduction in the rate of glucose absorption (Blackburn & Johnson 1980).

Katoch et al. (1971) suggested that the adverse effects of guar meal is due to decreased amino-acid absorption from the gut. Their studies showed an inhibtion of 27 to 53% in absorption of amino

acids *in vitro* due to the presence of 3% guar gum. The sulphur-containing amino acids seemed most severely affected and the likely effect of this may be to cause an imbalance in net availability of amino acids for protein synthesis.

The action of guar gum in reducing post-prandial glycaemia in man has also been demonstrated in other monogastric animals such as rats (Leeds *et al*. 1979) and pigs (Leeds & Kang 1980). Leeds *et al*. (1979) found that increasing meal viscosity slowed glucose disappearance from the gut and suggested that this was mainly due to prolonged gastric emptying. Johnson & Gee (1980) demonstrated *in vitro* that glucose transport was reduced in rat jejunum by the presence of 0.1, 0.25 and 0.5% guar gum. They suggested that this was due to an increase in the thickness of the unstirred layer at the mucosal surface, which itself was due to the high viscosity caused by the gum. Using rats, Blackburn & Johnson (1981) established an increase in the apparent viscosity of the small intestinal contents due to the ingestion of guar gum as a dry component of the food. No increase in viscosity was observed in the large intestine, presumably due to the degradation of guar gum by microflora. In a second experiment the rate of glucose absorption was decreased in jejunum segments pre-perfused with solutions of 0.5 and 0.6% guar gum, but the results were only significant when 0.6% was used.

Thus it appears that guar gum may influence the absorption of nutrients in at least two ways. The slowing of gastric emptying reduces the rate at which nutrients enter the small intestine and the apparent increased viscosity of the intestinal contents reduces absorption, perhaps by increasing the thickness of the unstirred layer and creating a physical barrier to the absorptive surfaces of the gut.

OBJECTIVES

In view of the ever-increasing demands for low-cost feedstuffs, the need for improved utilisation of conventional feedstuffs and/or the use of newer food sources, generally of low nutrient density, is becoming more important. A review of published reports clearly indicates the potential of dietary dilution by inert-additivies as a means of improving feedstuff utilisation; and the need to determine the tolerance of chicks for guar gum in order to enable the fullest use of guar meal to be made in poultry diets. The reports also indicate that the mechanism of action of guar gum in depressing chick growth is not fully understood.

This thesis comprises two section; 1. Dietary dilution by inert-additives, and 2. Studies on guar gum.

The principal objectives of the work on dietary dilution by inert-additives were to evaluate the published reports of dietary dilution improving chick performance, to determine the effect of inert additives upon the metabolisable energy values of conventional diets and to determine their mechanisms of action, if any. To meet these objectives:-

1. A variety of inert additives were studied in relation to their effect upon chick performance and the AME value of a typical broiler starter diet.
2. Two additives, sand and cellulose, were studied for their effect upon the TME value of a typical broiler starter diet.
3. Sand and cellulose were studied for their effect upon endogenous energy losses from cockerels.
4. The effect of sand and cellulose upon the size of the caeca of chicks was determined, and conventional and sand- and cellulose-diluted diets were used to evaluate the possible role of the caeca in digestion.
5. The role of the avian caeca with respect to endogenous energy loss was determined.
6. Sand was studied for its effect on long term growth of broilers and its mechanism of action was compared to that of low energy-dense diets.

The objectives of the studies on guar gum were to determine its tolerance level to chicks and to indicate its mechanism of action in

depressing chick growth by determining the effect of guar gum upon the ME value of a conventional diet, the digestibility of fat and nitrogen retention. Guar gum was:-

1. Included in a broiler diet at varying levels of inclusion to determine its effect on chick growth.
2. Studied for its effect upon the AME value of a conventional diet, the digestibility of fat and nitrogen retention.
3. Studied for its effect upon the TME value of a conventional diet.
4. Studied for its effect upon the endogenous energy losses of chicks.

Each experiment has been presented separately with a short discussion at the end of each. A general discussion, relating to all aspects, is presented at the end of each section.

Apparent Metabolisable Energy

The term Apparent Metabolisable Energy (AME) is a more correct definition of the metabolisable energy values devised by the system of Hill & Anderson (1958) and which have been widely used and accepted as a reliable basis for feed formulation (McNab, 1981). AME is the gross energy of the feed minus the energy lost as faeces, urine and combustible gases as a consequence of eating this feed (Harris, 1966). Gaseous losses from poultry are very small and usually considered to be negligible. In general, and in this thesis, AME values are corrected to zero nitrogen retention (AME_N) to allow for incomplete oxidation of retained nitrogenous components during catabolism.

Although used extensively over the past twenty years, AME determinations have been criticised for various reasons. AME determinations can vary between species (Fisher & Shannon, 1973; Sugden, 1974), strains (Sibbald & Slinger, 1963b; March & Biely, 1971) and animals of different ages (Renner & Hill, 1960; Lodhi et al. 1969; Rao & Clandinin, 1970). AME values are also dependent upon the level of feed intake (Guillaume & Summers, 1970). This is known to be due to the presence of endogenous energy losses (EEL's) which contribute to the gross energy content of the excreta.

Endogenous Energy Loss

The endogenous energy losses are considered to comprise metabolic faecal energy (FEm) plus endogenous urinary energy (UEe). When feed energy intake is high the EEL is relatively small but becomes of increasing significance when the energy intake is reduced, depressing the apparent ME value. Sibbald (1975) recognised the significance of this and devised a novel bioassay for measuring the True Metabolisable Energy (TME) content of feedstuffs (Sibbald, 1976).

The EEL can be determined by three different methods; with starved birds, with birds given an energy source which is completely absorbed and not excreted in the urine, e.g. glucose, or by extrapolation to zero intake of a line relating energy excretion to energy intake.

True Metabolisable Energy

The TME assay requires the determination of the metabolic faecal energy (FEm) and endogenous urinary energy (UEe). Sibbald (1976) stated that the correction of the conventional, or AME, values for EEL yielded true metabolisable energy (TME) values. He has also claimed that the TME determinations are subject to less variation caused by different feed intakes. In the TME assay the EEL is assumed to remain constant for the specific type and age of chicken used, irrespective of quantities or nature of the feed given to test birds.

Central to both the AME and TME systems is the assumption that there is a linear relationship between the energy voided as excreta and the energy input. The intercept value of the linear equation, i.e. the energy excretion at zero energy input, corresponds to the endogenous energy loss and is positive and non-zero (McNab, 1981). A relationship between AME and TME can be derived algebraically (Jonsson, 1980) and is as follows:-

$$AME = \frac{(TME \times FI) - EEL}{FI}$$

$$\text{or } AME = TME - EEL/FI$$

where AME and TME = energy values in kJ/g

FI = food intake in g

EEL = endogenous energy losses in kJ

Thus for a given constant TME value, AME is dependent upon the endogenous energy losses per unit of food intake. The dependency of AME values upon food intakes would be explained by variations in this ratio.

2. STUDIES ON DIETARY DILUTION

EXPERIMENT 2.1.

Objective

The experiment was conducted to determine the effect of dietary dilution by various inert fillers upon chick growth and feed conversion efficiency. The effect of the fillers on the Apparent Metabolisable Energy (AME) value of the diet was also studied.

Experimental

Design

Sixteen diets were fed to single-sexed broiler chicks from 7 to 21 days of age. The diets were randomly allocated to two groups of three birds in each of three blocks consisting of 16 double cages per block (one double cage per treatment per block). Five inert fillers were included at each of three dietary inclusion concentrations with six replicate groups of chicks receiving each diet. A control diet, i.e. one containing no filler, was also fed to six replicate groups.

Birds and Management

About 400 P.R.C. stock male broiler chicks from the same hatch were wing-banded and reared to 7 days of age in thermostatically controlled battery brooders. During the rearing period the chicks were fed a conventional starter mash which was also used as the basal diet during the experimental period (Table 2.1).

At 7 days of age the chicks were individually weighed; 288 chicks were selected from the middle weight band and distributed to one of forty-eight double cages, according to a previously randomised plan, so that each half-cage contained 3 chicks. Each double cage was divided into two by a mesh partition, each half having its own food trough but with a shared water trough and a continuous raised floor. The cages were all located in the same room in which the environment was controlled. Food and drinking water were available *ad lib.* at all times. Total food intake for

each group of 3 birds was recorded for the 14 day period and the birds were again individually weighed at the end of the experiment, i.e. at 21 days of age.

During the last four days of the experiment an **AME assay** was carried out. The wire floors of the cages were cleaned and a clean tray placed under each cage. All excreta from each group of six birds were collected quantitatively at the end of the four day period and total food intake for each group of six birds was recorded. The excreta were oven-dried, allowed to equilibrate to atmospheric moisture, weighed and analysed for gross energy and total nitrogen content for the determination of Apparent Metabolisable Energy.

Diets

The composition of the basal diet (diet 1) is described in Table 2.1. The remaining 15 experimental diets consisted of the basal diet diluted by weight by the inclusion at 20, 40, and 60 g/kg, respectively, of the following fillers:-

Sand	:	diets 2, 3 & 4
Cellulose	:	" 5, 6 & 7
Kaolin	:	" 8, 9 & 10
Sawdust	:	" 11, 12 & 13
Cement Kiln dust	:	" 14, 15 & 16

The diets were presented in mash form. All diets were analysed for dry matter (DM), gross energy and total nitrogen.

Results

The results are summarised in Table 2.2. Because it is assumed that none of the diluents used in this experiment are metabolised by chicks food intakes were adjusted to give intakes of basal diet, i.e. total intake less diluent. The AME values of the diets are expressed as MJ/kg DM basal diet and are corrected to zero nitrogen retention. The term Ein - Eout refers to the difference between the energy intake in the food and the energy output in the excreta. No account was taken of the contribution to the gross

Table 2.1. Composition of diet 1

Ingredient	g/kg
Herring meal (CP=74%)	100
Maize	350
Wheat	183
Barley	75
Soya bean meal (CP=44%)	180
Meat & Bone meal (CP=51%)	40
Maize oil	40
$CaCO_3$	5
$CaHPO_4$	18
NaCl	2
DL-Methionine	2
Mineral premix[1]	2.5
Vitamin premix[2]	2.5

1. Providing per kg diet; 3.5mg Cu,0.4mg I,80mg Fe, 300mg Mg,100mg Mn,50mg Zn.

2. Providing per kg diet; 2000 i.u. Vitamin A,600 i.u. Vitamin D_3,25mg Vitamin E,1.3mg menaphthone,4mg riboflavin, 28mg nicotinic acid,10mg pantothenic acid,0.05mg biotin.

Table 2.2 Mean body weight gain, basal food intake, fce, and results of AME study on chicks receiving diets diluted with inert fillers.

Diet; level g/kg		Body wt. gain g	Food intake (FI) g	fce	FI over AME study g	Ein-Eout MJ	AME MJ/kg	$\hat{TME}$ MJ/kg
Control		387	611	.63	84	1.310	15.63	12.30
Sand;	20	353	581	.61	95	1.506	15.83	21.05
	40	378	595	.64	85	1.317	15.55	16.82
	60	362	559	.65	80	1.258	15.64	16.20
Cellulose;	20	351	577	.61	82	1.254	15.39	-1.91
	40	358	579	.62	81	1.256	15.50	13.21
	60	317	531	.60	75	1.192	15.92	15.68
Kaolin;	20	356	583	.61	70	1.048	14.95	18.43
	40	362	585	.62	76	1.177	15.40	16.53
	60	330	530	.62	59	.873	14.74	14.94
Sawdust;	20	336	562	.60	82	1.237	15.17	19.47
	40	353	588	.60	82	1.260	15.32	15.90
	60	347	561	.62	85	1.334	15.59	29.60
CKD;	20	332	566	.59	74	1.092	14.76	15.17
	40	332	542	.61	80	1.271	15.81	15.78
	60	320	563	.57	70	1.056	15.04	21.75

energy of the excreta by either cellulose or sawdust. The TME values of the diets were estimated from the relationship:-

$$TME = \frac{(Ein-Eout) + EEL}{FI}$$

where Ein = energy intake
Eout= energy in excreta
EEL = endogenous energy loss
FI = basal food intake

Standard errors of the means may be estimated from the mean squares (MS) from the Analysis of Variance tables in the Appendix.

Birds receiving the control diet were statistically significantly heavier than all other groups of birds ($p<0.01$). Of the birds receiving diluted diets those receiving the sand-diluted diets had the highest body weight gain, those receiving CKD the lowest. The weight gains of the remaining birds were very similar and lay between the extremes of sand and CKD. Although there appears to be a significant effect of the level of diluent ($p<0.05$), the pattern is not one of a steady decline but indeed increases at the 40g/kg level.

The pattern of response of basal food intakes and fce are much the same as that of body weight gains, with the control group of birds consuming more food than the other groups of birds ($p<0.05$) and having a higher fce although this was not found to be statistically significant.

During the AME study the basal food intakes of the different groups of birds were significantly different ($p<0.01$), and this was reflected in the term Ein-Eout ($p<0.001$). Thus there also appears to be significant differences in the AME values of the diluted diets ($p<0.01$). There is,however, no significant difference between the AME of the control diet and those of the diluted diets.

The analysis of variance of the estimated TME values suggests that there is a significant difference

between the $\hat{TME}$ values of each of the diets and also that each group of birds experienced a different endogenous energy loss. With the exception of cellulose at 20g/kg the $\hat{TME}$ values for all treatments exceed that of the control diet. There is, however, no statistically significant difference between the $\hat{TME}$ of the control diet and the mean $\hat{TME}$ of all the other treatments. The greatest treatment mean (i.e. the mean value of the three levels) being for the sand-diluted diets. The low, negative value for the diet containing 20 g cellulose/kg is due to the problem of regressing three very similar points.

Discussion

The results of the AME study and the estimated TME values support further studies on dietary dilution by inert additives. The differences between the AME values of the diluted diets are a function of the differences in food intakes; AME values are known to be dependent upon the level of feed intake (Guillaume & Summers, 1970). The estimated TME values suggest that dietary dilution improves the energy availability of a diet although the difference between the estimated TME of the control diet and the mean values of the other treatments is not statistically significant.

Body weight gains are, however, significantly reduced by dietary dilution. This would support the observation by Hooge (1979) that, with sand-diluted diets, broilers require slightly longer to reach a given weight. Since the fce of the control group of birds was not statistically greater than that of the other treatments, dietary dilution could still prove to be economical despite the reduced body weight gains.

In terms of body weight gains, fce and estimated TME sand would appear to be the most suitable diluent for use in broiler diets. Body weight gains were reduced

by a considerably greater extent by the other diluents which would limit their suitability.

Birds receiving kaolin-diluted diets show a greater reduction in body weight gains and feed intake at the highest level of inclusion (60g/kg) which suggests that the bulky nature of kaolin limited the amount of feed that could be ingested.

Cement kiln dust-diluted diets resulted in the most severe reduction in body weight gains, indicating its unsuitability for use as a diluent in broiler diets. Within the treatment food intakes varied little and thus the reduction in body weight gain was probably not only a function of food intake and may have resulted from a possible toxicity of the CKD.

Cellulose-diluted diets had the lowest estimated TME values of the diluted diets and as such were the most comparable to the control diet. The most severe reduction in body weight gain and feed intake was at the 60g cellulose/kg level of inclusion. Combined with an estimated TME that is not statistically different from that of the control diet this suggests that the depression in growth is the result of a failure to consume sufficient feed to satistfy requirements, probably due to the bulky nature of cellulose.

The estimated TME values of the sawdust-diluted diets were most comparable to those of the sand-diluted diets, but body weight gains were considerably less. The treatment means of food intakes of sawdust- and sand-diluted diets are not greatly different (570 and 578g respectively) which suggests a less efficient use of dietary energy from sawdust-diluted diets. Energy expenditure of the birds may have been increased by the presence of sawdust, for example, the bulky nature of the diets may have increased energy requirements for the processes of ingestion and digestion.

EXPERIMENT 2.2.

Objective

This experiment was designed to confirm the estimated TME values for sand- and cellulose-diluted diets obtained in Experiment 2.1 by measuring the TME directly, using the rapid bioassay method of Sibbald (1976).

Experimental

Design

The experimental design was that of a randomised block with six replicates of eight dietary treatments which were as follows:-

1. diet 1 - basal diet (control)
2. diet 2 - 20 g sand/kg basal diet
3. diet 3 - 40 g " "
4. diet 4 - 60 g " "
5. diet 5 - 20 g cellulose/kg basal diet
6. diet 6 - 40 g " "
7. diet 7 - 60 g " "
8. starved control birds.

Birds and Management

Forty-eight dubbed, adult White Leghorn cockerels which had never had access to grit were fasted for 24 hours prior to being fed 25 g of test diet through a tube, the end of which was placed just at the opening to the birds' crop. After the feeding, each bird was placed in a clean wire cage, a clean plastic tray placed underneath and the time of housing recorded. The excreta voided in the subsequent 24 hours were quantitatively collected, frozen and then freeze-dried. Six birds which remained fasted were also maintained for the collection of endogenous losses. Drinking water was available ad lib. at all times.

The freeze-dried samples were equilibrated to atmospheric moisture for 24 hours, weighed and then analysed for gross energy

and total nitrogen content for the determination of TME_N.

Prior to the experimental period all birds were maintained on a holding diet of the following composition (g/kg): maize meal - 743, soya bean meal - 174, maize oil - 20, pruteen* - 20, limestone - 15, dicalcium phosphate - 20, sodium chloride - 3, vitamin and mineral supplement - 5.

Diets

The diets used during the experimental period were samples taken from the control and the sand- and cellulose-diluted diets used in Experiment 2.1.

Results

The TME values were calculated using the following equation:-

$$TME = \frac{Ein - (Eout - EEL)}{FI}$$

where Ein = gross energy intake, MJ
Eout = gross energy of excreta, MJ
EEL = endogenous energy loss, MJ
FI = food input, kg

The EEL was taken as the mean value of the gross energy of the output from the starved birds and was 51.17 kJ/24 hours. It was assumed that neither sand nor cellulose were metabolised by poultry and accordingly the food input was expressed as basal food input, i.e. 25 g x (100 - % dilution). TME values are expressed in terms of MJ/kg DM basal diet and are corrected to zero nitrogen retention (TME_N).

Sand

Table 2.3 shows the mean TME_N values of the sand-diluted diets. Despite a numerical increase there were no statistically significant differences between the TME_N values of the four diets.

Cellulose

The results of the TME_N determination in Table 2.4 show no statistical differences between the four cellulose-diluted diets.

* See page 38.

Table 2.3. TME_N of Sand-diluted diets

Diet	1	2	3	4	
Sand g/kg	0	20	40	60	
Mean TME_N MJ/kg DM	16.94	17.18	17.43	16.97	NS
	±SE .24	±SE .24	±SE .24	±SE .26	

Table 2.4 TME_N of Cellulose-diluted diets

Diet	1	5	6	7	
Cellulose g/kg	0	20	40	60	
Mean TME_N MJ/kg DM	16.94	16.89	16.65	17.23	NS
	±SE .15	±SE .18	±SE .16	±SE .18	

NS = not significant

Discussion

Sand

At first sight these data do not confirm the increase in TME_N as a result of including sand in diets as suggested in Experiment 2.1. The calculation of the TME_N value of these diets assumed a value for EEL estimated from the mean of those excreted by six starved control birds. If, however, the EEL was influenced by the nature of the diet this could affect the calculation of TME. For example, if the abrasive nature of the sand were to increase the amount of endogenous energy lost then the TME of the diets containing sand would be greater than the values calculated above and a pattern similar to that suggested in Experiment 2.1 would be observed.

It is also possible that the exposure to the sand over a period of time during Experiment 2.1 increased the birds capacity to absorb and metabolise energy, thereby increasing the TME. The presence of sand may in some way increase the surface area available for absorption, perhaps by stimulating villi growth. Stimulation of the general growth of the gut or part of it by sand cannot be ruled out.

Cellulose

The results confirm the findings of Experiment 2.1, in that there are no statistically significant differences between the TME_N values of the basal diet portions of the four diets. There is again an increase in the TME value of the diet containing 60g cellulose/kg basal diet. There may, therefore, be an effect of cellulose upon TME but it is likely to be only slight and unable to be detected statistically by the experimental design and/or degree of replication used here.

*Pruteen is a protein-rich (CP=72%) product marketed by ICI. It is produced by the organism Methylophilus methylotropino which grows rapidly on a medium of methanol, ammonia and oxygen.

EXPERIMENT 2.3.

Objective

The aim of this experiment was to determine the effect of the passage of sand upon endogenous energy losses from adult cockerels.

Experimental

Design

The design was that of a randomised block involving six replicates of four treatments which were inputs of 0, 12.5, 25.0 and 50.0 g of sand.

Birds and Management

The procedure followed was basically that described for Experiment 2.2 but included modifications to the original Sibbald method.

Twenty-four dubbed, adult Warren cockerels that had never had access to grit were fasted for 48 hours during which time they were twice tube-fed 50 ml of a 500 g/kg glucose solution, approximately 6 and 30 hours after housing. After the starvation period the birds were tube-fed a known quantity of sand according to a previously randomised plan, returned to clean cages with a clean tray placed underneath and the time of housing recorded. The control birds received no sand and were tube-fed 50 g granulated glucose. About 30 hours after feeding all birds were tube-fed 50 ml water. The excreta voided during the 48 hours subsequent to feeding were collected quantitatively, frozen and then freeze-dried. Drinking water was available ad lib. at all times.

The freeze-dried samples were allowed to equilibrate to atmospheric moisture, weighed and then analysed for gross energy and total nitrogen content.

Prior to the experiment the birds were maintained on a holding diet, the composition of which is given in the Experimental section in Experiment 2.2.

Table 2.5. The effect of sand upon mean endogenous energy loss

Sand g	0	12.5	25.0	50.0		
EEL kJ/48h	73.78	73.32	76.99	62.43	±SE 6.51	NS
EEL_N kJ/48h	35.03	38.35	37.53	29.33	±SE 4.47	NS

NS = not significant

Results

The mean endogenous energy losses of the birds fed the four levels of sand are given in Table 2.5. The energy losses are expressed in kilojoules (kJ) per 48 hours and are shown both before (EEL) and after correction to zero nitrogen retention (EEL_N).

Despite some numerical variations the sand had no significant effect upon the endogenous energy losses from the birds.

Discussion

It does not appear from the data obtained in this experiment that the discrepancy between the estimated TME values of sand-diluted diets in Experiment 2.1, which were significantly increased, and the TME values of the same diets measured in Experiment 2.2, which showed no significant differences, was due to an increase in the endogenous energy loss.

The EEL has both faecal and urinary components (McNab, 1981). The faecal components are unabsorbed bile, digestive juices and intestinal mucosal cells and the urinary component is thought to consist mainly of waste products of nitrogen catabolism. It might be expected that the abrasive nature of sand would increase the sloughing process of the mucosal cells thereby increasing the faecal component of the EEL. It is possible, however, that the mucosal cells constitute such a small part of the endogenous loss that any increase would be diluted by other components and be difficult to detect.

EXPERIMENT 2.4.

Objective

This study was carried out in order to determine whether or not the inclusion of either sand or cellulose in a practical diet has any effect on the size of the caeca in domestic fowl.

Experimental

Design

Three treatments were randomly assigned to 30 chicks, giving 10 replicates of each. The treatments were:-

1. diet 1. - basal diet (control)
2. diet 2. - 60 g sand/kg basal diet
3. diet 3. - 60 g cellulose/kg basal diet

Birds and Management

About 50 day-old Marshalls' stock, male broiler chicks from the same hatch were wing-banded and reared to 7 days of age in thermostatically controlled battery brooders, during which time they were fed a broiler starter mash, the composition of which is given in Table 2.1, Experiment 2.1. At 7 days of age the chicks were individually weighed; 30 chicks were selected from the middle weight band and randomly assigned to the three treatments.

The chicks remained housed in the battery brooders. At 21 days of age the chicks were killed by an intravenous injection of Nembutal (pentobarbitone sodium, 60 mg/ml). The caeca were dissected out and individually measured lengthwise, care being taken that they were not stretched.

Diets

Diet 1, the control diet, was formulated to the same composition as the control diet in Experiment 2.1 (Table 2.1). The diluted diets consisted of the control diet diluted by weight by 60 g/kg of sand or cellulose, diets 2 and 3 respectively.

Table 2.6 The length of caeca of chicks reared on sand and cellulose diluted diets.

Diet	n	Mean body weight g	Length of caeca, mm Left	Right
Control	10	466 ±SE 10.5	106.3	98.0
60g sand/kg	10	462 ±SE 2.3	109.8	102.5
60g cellulose/kg	10	460 ±SE 1.1	109.5	103.8
		NS	±SE 3.84^{NS}	4.04^{NS}

NS = not significant

Results

The lengths of the caeca of birds fed sand- and cellulose-diluted diets are presented in Table 2.6. Analysis of variance shows no significant differences between the lengths of the caeca on the three treatments although there are numerical increases in the lengths of the caeca of the birds fed both the sand- and cellulose-diluted diet, compared to the birds fed the control diet. The numerical differences between the lengths of the left and right caeca are also not statistically significant.

Discussion

Although there are no statistically significant differences there does appear to be a trend towards larger caeca in the birds fed sand- and cellulose-diluted diets. It is possible that the population size used in this experiment was too small to detect accurately a difference in the caecal dimensions.

An increase or change in the size of the caeca could indicate a larger, or different, microbial population within them which could improve the metabolisable energy of the diet by further digestion of the residues within the caeca. This is perhaps more conceivable in the case of the cellulose-diluted diets since it has been suggested that at least some of the crude fibre eaten by chickens is digested in the caeca (Thornburn & Willcox, 1965a). This would not necessarily explain any effect of sand, but it is possible that an increase in the microbial population due to the presence of sand could itself be the stimulant of increased caecal growth.

EXPERIMENT 2.5.

Objective

This experiment was designed to investigate whether birds with caeca metabolise more of an undiluted conventional diet or one diluted with either sand or cellulose than birds without caeca. The TME was calculated from the slope of a line derived from linear regression of three intake levels of each diet.

Experimental

Design

Three diets were randomly assigned to two groups of birds, intact and caecectomised, and were fed at three input levels of intake - ad lib., 40 g and 20 g. **Each group comprised 14 birds.**

Birds and management

Male, White Leghorn chicks were reared from day old to 10 weeks of age in thermostatically controlled battery brooders, on a typical chick starter diet. At 10 weeks of age half the birds were caecectomised by anaesthetising them with Penthrane (Methoxyflurane BP) and surgically removing the caeca. After regaining conciousness the birds were returned to a clean battery brooder. Recovery was observed by daily weight measurement of the birds until their body weights were equivalent to those of unoperated control birds.

At 16 weeks of age the intact and caecectomised birds were removed to individual cages and the diet changed to a typical layers' mash. After daily feed intakes had stabilised the birds were put on to a meal feeding regime, in which they were trained to eat their daily food intakes in two hours per day. Intakes were recorded and when they had stabilised at a maximum the birds were randomly assigned to the first set of experimental diets, which were cellulose-diluted. After a ten-day adaptation period the first experiment was carried out at 21 weeks of age. The birds were starved for 48 hours and then allowed to eat ad lib. for 2 hours. Clean trays were placed

under the cages and the excreta voided over the following 48 hours were collected. The same procedure was repeated with intakes of 40 g and 20 g.

A second set of experimental diets, which were sand-diluted, were then randomised amongst the birds and a 10-day adaptation period followed. At 24 weeks of age the experiment was repeated.

Excreta samples were oven-dried and allowed to equilibrate to atmospheric moisture, weighed and analysed for gross energy and total nitrogen.

Diets

The composition of the chick starter diet and the layers' diet are presented in Tables 2.7 and 2.8 respectively.

The two sets of experimental diets consisted of the layers' diet (diet 1) diluted by weight with 40 g/kg and 60 g/kg of either sand or cellulose.

The experimental diets were, therefore:-

Set 1 - cellulose diluted
Diet 1 - basal diet (control)
Diet 2 - 40 g cellulose/kg basal diet
Diet 3 - 60 g cellulose/kg basal diet

Set 2 - sand diluted
Diet 1 - basal diet (control)
Diet 4 - 40 g sand/kg basal diet
Diet 5 - 60 g sand/kg basal diet

The diets were analysed for dry matter, gross energy and total nitrogen.

Results

The results were treated in the following manner. Food intakes were corrected for the presence of diluent (i.e. total intake less diluent). Energy balance was corrected to zero nitrogen retention and the derived TME_N values are presented in terms of MJ/kg DM.

An analytical model was designed incorporating the diet, the presence or absence of caeca, food intake and energy balance. The

Table 2.7 Composition of PRC chick starter diet

Ingredients	g/kg
Barley	100
Maize	300
Wheat	245
Herring meal (CP=74%)	50
Soya bean meal (CP= 44%)	220
Grass	50
Limestone	5.3
Dicalcium phosphate	21.7
Vitamin premix 4[1]	2.5
Mineral premix 5[2]	2.5
NaCl	2.5
Pancoxin*	0.454

1. Providing per kg diet; 2000 i.u. Vitamin A, 600 i.u. Vitamin D , 25mg Vitamin E, 1.3mg menaphthone, 4mg riboflavin, 28mg nicotinic acid, 10mg pantothenic acid, 0.05mg biotin.

2. Providing per kg diet; 3.5mg Cu, 0.4mg I, 80mg Fe, 300mg Mg, 100mg Mn, 50mg Zn.

* Coccidiostat

Table 2.8 Composition of PRC Layer's diet

Ingredients	g/kg
Barley	255
Maize	242
Wheat	193
Herring meal (CP=74%)	48
Soya bean meal (CP=44%)	104
Grass meal	50
Meat & Bone meal (CP=51%)	19
Limestone	68
Dicalcium phosphate	14
Vitamin premix 3[1]	2.5
Mineral premix 5[2]	2.5
NaCl	2.0

1. Providing per kg diet; 6000 i.u. Vitamin A, 800 i.u. Vitamin D , 25mg Vitamin E, 1.3mg menaphthone, 4mg riboflavin, 28mg nicotinic acid, 10mg pantothenic acid.

2. Providing per kg diet; 3.5mg Cu, 0.4mg I, 80mg Fe, 300mg Mg, 100mg Mn, 50mg Zn.

effect of caecectomy was determined by the removal of the caecal parameter from the analytical model. The TME_N was estimated by plotting energy balance against intake. To determine whether or not there were any differences between the TME_N value of the three diets a model was prepared involving a single slope and intercept for all diets, i.e. such that all diets had the same TME_N value and the birds had the same EEL_N, much in the same way as in Experiment 2.1. This model was then compared to two others, one assuming that the diets had the same TME_N but that the birds had differences in EEL_N (i.e. several intercepts, same slope) and the other assuming that the diets had different TME_N values but that the birds had the same EEL_N (i.e. several slopes, single intercept).

Cellulose

There did not appear to be any effect of caecectomy on the TME_N values of the diets. Further analysis was then carried out on combined, intact and caecectomised, results. Table 2.9 gives the estimated TME_N values of the cellulose diluted diets. There was a significant depression in the TME_N value of the diet due to the inclusion of cellulose ($p<0.05$). There was not, however, any effect on the EEL_N.

Sand

There was no effect of caecectomy on the TME_N values of the diets and so, again, the two sets of results were combined. The TME_N values of the sand-diluted diets proved difficult to interpret. Both the model involving different TME_N's but the same EEL (several slopes, single intercept) and the one involving different EEL's but the same TME_N (single slope, several intercepts) proved a better fit than the single slope and intercept model, but neither proved to be a better fit than the other. Thus it is possible that the presence of sand either improved the TME_N of the diet whilst having no effect on the EEL_N (Table 2.10a) or had no effect on the TME_N but did effect the EEL_N (Table 2.10b).

Table 2. TME_N values of cellulose-diluted diets obtained by linear regression of energy balance at three levels of intake

Diet	1	2	3	
Cellulose g/kg	0	40	60	
TME_N MJ/kg DM	11.88 ±SE .32	10.48 ±SE .39	10.54 ±SE .39	*
EEL_N kJ/48 hours	_____	53.89	_____	

* = $p<0.05$

Table 2.10. Proposed TME_N values of sand-diluted diets obtained by linear regression of energy balance at three levels of intake

a. Diets having different TME_N's but the birds having the same EEL_N

Diet	1	4	5
Sand g/kg	0	40	60
TME_N MJ/kg DM	12.23 ±SE .27	11.26 ±SE .29	12.75 ±SE .27
EEL_N kJ/48 hours	———	60.26	———

b. Diets having the same TME_N but the birds having different EEL_N's

Diet	1	4	5
Sand g/kg	0	40	60
TME_N MJ/kg DM	———	12.13 ±SE.23	———
EEL_N kJ/48 hours	52.39	100.83	33.21

Discussion

The lack of effect of caecectomy on the TME values of the control, sand - and cellulose - diluted diets confirms the opinion that the caeca do not play an important role in digestion (McNab, 1973; Nakahiro *et al*. 1974; Nakahiro & Isshiki, 1975), although there have been reports of crude fibre digestion within the caeca (Mangold, 1934; Halnan, 1949; Thornburn & Willcox, 1965a). It is possible that even in the cellulose diluted diets any possible effect on the crude fibre fraction could have been diluted by the high digestibility of the remainder of the diet and not detected by the method used here.

Cellulose

In previous experiments reported in this thesis the presence of cellulose in the diet has had no effect on AME_N, estimated TME (Experiment 2.1) or measured TME_N (Experiment 2.2). However, in this experiment there is a significant depression in TME_N. Since the food intakes used had been corrected for the presence of diluent the depression cannot be considered to be due to the dilution. Furthermore, the effects observed in this experiment are greater than would be expected due to dilution (expected values would be:- 40 g/kg cellulose - 11.40 MJ/kg; 60 g/kg cellulose - 11.17 MJ/kg).

In both Experiment 2.1 and 2.2, although there was a numerical decrease in AME and TME at the 40 g/kg inclusion level this was not considered significant. There appears, therefore, to be no consistent pattern in the three experiments. Replications in the three experiments were 3, 6 and 4 respectively and so any difference in results is not due to a larger replication allowing a small effect to be detected. It is also unlikely that the difference in experimental methods used here would account for the difference in results; essentially the method is the same as that used in the AME assay (Experiment 2.1).

The inconsistency of the results compared with those obtained previously, coupled with the fact that the probability of observing at least one significant result with a probability >0.05 in three experiments when there is no true difference is 0.14, makes it seem likely that the effects observed are as a result of chance.

Sand

Little can be concluded from the results here without considering previous data. The results from Experiment 2.1 suggested that sand tended to increase the estimated dietary TME value but this conclusion was not confirmed by the measurement of TME_N in Experiment 2.2. However, in Experiment 2.3 it was shown that sand has no effect upon EEL_N. Taking these results into consideration it is more likely that the results presented in Table 2.10a reflect the true effect of sand upon the ME of diets; i.e. the TME_N is increased whilst there is no effect on the EEL_N.

EXPERIMENT 2.6.

Objective

This experiment was carried out to determine whether or not poultry are able to digest cellulose and, if so, whether or not this ability is affected by caecectomy.

Experimental

Design

Three input levels of cellulose were randomly assigned to each of two groups of eleven birds, intact and caecectomised. There were four replicates in each group for input levels 1 and 2, and three replicates for level 3.

Birds and Management

Twenty-two dubbed adult White Leghorn cockerels, eleven each of intact and caecectomised birds, were housed in individual cages and starved for 48 hours. During the starvation period each bird twice received, at 6 hours and 30 hours, 50 ml of a 500 g/kg glucose solution. The experimental method followed was that described in Experiment 2.3. After the starvation period the birds were tube-fed 10, 20 or 30 g pelleted cellulose, according to a previously randomised plan, returned to clean cages and the time of housing recorded. About 30 hours after feeding the birds were tube-fed 50 ml water. The excreta voided over the 48 hours subsequent to feeding were quantitatively collected. The excreta samples were freeze-dried and analysed for gross energy and total nitrogen.

Results

The energy balance figures for the three input levels of cellulose are presented in Table 2.11. The energy balance figures are all negative and essentially represent the energy losses from

Table 2.11. Mean energy balance of intact and caecectomised birds fed three input levels of cellulose

	Energy Balance (Ein - Eout) kJ/48h				Energy Balance corrected to zero N_2 retention kJ/48h			
Cellulose (g)	10	20	30		10	20	30	
Intact birds	-94.20	-101.78	-120.01	NS	-51.27	-50.23	-54.39	NS
	±SE 7.3	±SE 7.3	±SE 8.43		±SE 5.42	±SE 5.42	±SE 6.26	
Caecectomised	-89.49	-119.70	-132.72	*	-49.98	-71.44	-69.4o	*
	±SE 9.28	±SE 9.28	±SE 10.71		±SE 4.01	±SE 4.01	±SE 4.63	
	NS	NS	NS		NS	NS	NS	

NS = not significant

* = $p<0.05$

the birds. The figures are in kJ/48 hours and are presented both before and after correction to zero nitrogen retention.

In the case of intact birds there were no significant differences between the energy losses of the birds receiving the three different input levels of cellulose. Correcting the figures to zero nitrogen retention did not affect this conclusion. With the caecectomised birds, however, there was a significant increase in energy loss with increasing intake of cellulose, which remained after nitrogen correction. There appeared to be no effect of caecectomy on the energy losses.

Discussion

The results suggest that no digestion of cellulose occurs within the digestive tract of poultry, not even within the caeca. The increase in energy loss within the caecectomised birds could be due to the increased water loss experienced by these birds. The water would have been absorbed by the cellulose, increasing its' bulk further and this may have caused greater surface erosion throughout the gut, thereby increasing the faecal component of the endogenous energy losses.

EXPERIMENT 2.7.

Objective

This experiment was designed to determine whether the effects observed when sand is included in diets for poultry are caused by its' acting as a diluent only, such that equivalent results would be obtained by using diets of reduced energy and crude protein content. The effect of low dietary concentrations of sand upon broiler growth and food conversion efficiency (fce) was studied and compared to the effect of lower energy and protein levels than those present in a conventional broiler diet.

Experimental

Design

Forty-eight groups, each of 40 male or female broiler chicks, were randomly assigned to receive one of eight experimental diets from day-old to 56 days of age. Each diet was randomly assigned to 3 pens of both male and female birds. The experiment was of a factorial design where the factors were 2 treatments x 4 levels x 2 sexes x 3 replicates. The two treatments were dietary dilution by sand and "dilution" by energy (ME) and protein (CP) reduction. The four levels were:

- for sand dilution - 0, 20, 40 and 60 g sand/kg diet
- for ME and CP reduction - 100, 98, 96 and 94% of the energy and crude protein content of the control diet.

Birds and management

About nineteen hundred PRC stock broiler chicks were floor-brooded in 48 pens under one roof and in a controlled environment. The pens had a layer of wood shavings on the floor. Infra-red brooding lamps were provided for the first 7 day period. A floor area of about 0.09 m^2 per bird was allowed. Male and female chicks were

allocated randomly, at one day of age, to the separate pens where food and drinking water were available ad lib. at all times.

Total pen weights were obtained by weighing groups of birds at 4 and 8 weeks of age. Food eaten per pen was recorded weekly. Mortality was also recorded.

Diets

The composition of the experimental diets was changed after 28 days to allow for the different energy and protein requirements during the period of 28-56 days. All diets were presented in pellet form.

The composition of the starter (0-28 days) and finisher (28-56 days) diets are presented in Tables 2.12 and 2.13 respectively. The control starter diet was of the same composition as the control diet used in previous experiments. The reduced energy and protein diets were formulated to contain 98, 96 and 94% of both the energy and crude protein of the control diet, i.e. that contained in the sand-diluted diets.

The TME_N values of the diets were determined by the Sibbald bioassay with the modifications devised at the PRC already mentioned in this thesis. Both the starvation and collection periods were 48 hours. During the starvation period all birds were twice tube-fed 50 ml glucose solution (500 g/kg) and on feeding the starved control birds were tube-fed 50 g granulated glucose. All birds received 50 ml water about 30 hours after feeding. Excreta samples were treated in the usual manner.

The diets were isonitrogenous and the amino acids balanced.

Results

Analysis of variance was carried out using Genstat (1980, Lawes Agricultural Trust, (Rothampsted Experimental Station)). The analyses were on a logarithmic scale.

Table 2.14 and 2.15 summarise the performance of birds on sand-diluted diets at 4 and 8 weeks respectively. Likewise, Tables 2.16 and 2.17 summarise the performance of birds on ME and CP-reduced diets. In the case of sand-diluted diets adjusted food intakes and fce (i.e. whole diet less diluent) is also given. Standard errors

Table 2.12. Composition of starter diet (0 - 28 days) g/kg

Diet	1	2	3	4	5	6	7	8
	Controls		Sand-diluted			Energy, protein reduced		
Barley	75.0		73.5	72.0	70.5	115.0	180.0	185.0
Maize	350.0		343.0	336.0	329.0	302.0	250.0	220.0
Wheat	183.0		179.34	175.68	172.02	183.0	183.0	183.0
Herring meal (CP = 74%)	100.0		98.0	96.0	94.0	75.0	65.0	25.0
Soya bean meal (CP = 44%)	180.0		176.4	172.8	169.2	210.0	210.0	270.0
Meat & bone meal (CP = 51%)	40.0		39.2	38.4	37.6	40.0	40.0	45.0
Maize oil	40.0		39.2	38.4	37.6	40.0	35.0	35.0
Limestone	5.0		4.9	4.8	4.7	5.0	5.0	5.0
Dicalcium phosphate	18.0		17.64	17.28	16.92	18.0	18.0	18.0
DL-Methionine	2.0		1.96	1.92	1.88	5.0	7.0	7.0
Vitamin premix 4*	2.5		2.46	2.40	2.36	2.5	2.5	2.5
Mineral premix 5*	2.5		2.46	2.40	2.36	2.5	2.5	2.5
NaCl	2.0		1.96	1.92	1.88	2.0	2.0	2.0
Builders sand			20.0	40.0	60.0			
Calculated ME MJ/kg	12.68		12.43	12.17	11.90	12.43	12.14	11.92
Calculated crude protein %	23.04		22.57	22.12	21.67	22.57	22.13	21.88

* FOR COMPOSITION PER Kg DIET SEE TABLE 2.12a

Table 2.12a. Vitamin and Mineral composition per kg diet.

Diet	1A,2A,6A, 7A & 8A	3A	4A	5A
Vitamin				
A, i.u.	2000	1968	1920	1888
D_3, i.u.	600.0	590.4	576.0	566.4
E, mg	25.0	24.6	24.0	23.6
Menaphthone, mg	1.3	1.28	1.25	1.23
Riboflavin, mg	4.0	3.94	3.84	3.78
Nicotinic acid, mg	28.0	27.55	26.88	26.43
Pantothenic acid, mg	10.0	9.84	9.60	9.44
Biotin, mg	0.05	0.049	0.048	0.047
Mineral				
Cu, mg	3.5	3.44	3.36	3.30
I, mg	0.4	0.394	0.384	0.378
Fe, mg	80.0	78.72	76.80	75.52
Mg, mg	300.0	295.2	288.0	283.2
Mn, mg	100.0	98.4	96.0	94.4
Zn, mg	50.0	49.2	48.0	47.2

Table 2.13. Composition of finisher diets (28-56 days) g/kg

Diet	1A	2A	3A	4A	5A	6A	7A	8A
	CONTROLS		SAND DILUTED			ENERGY, PROTEIN REDUCED		
Barley							239.0	302.0
Maize	141.0		138.0	135.0	132.54	141.0	70.0	31.2
Wheat	530.0		519.0	508.8	498.20	551.0	394.0	387.0
Herring meal (CP = 74%)	37.6		36.85	36.10	35.34	15.0	14.5	14.6
Soya bean meal (CP = 44%)	248.0		243.0	238.1	233.12	253.0	238.0	220.0
Maize oil	15.04		14.74	14.44	14.14	11.0	14.5	11.0
Limestone	9.40		9.21	9.02	8.84	10.0	10.0	10.0
Dicalcium phosphate	9.40		9.21	9.02	8.84	10.0	10.0	10.0
Choline chloride	0.3		0.29	0.29	0.28	0.3	0.3	0.3
Vitamin premix 4*	2.5		2.45	2.4	2.35	2.5	2.5	2.5
Mineral premix 5*	2.5		2.45	2.4	2.35	2.5	2.5	2.5
NaCl	2.0		1.96	1.94	1.88	2.0	2.0	2.0
L-lysine	1.5		1.47	1.44	1.41	1.6	1.6	1.6
Builders sand			20.0	40.0	60.0			
Calculated ME MJ/ka	11.86		11.62	11.39	11.15	11.59	11.43	11.19
Calculated crude protein %	18.9		18.52	18.14	17.77	18.52	18.14	17.83

* FOR COMPOSITION PER Kg DIET SEE TABLE 2.13a

Table 2.13a. Vitamin and Mineral composition per kg diet.

Diet	1A,2A,6A 7A & 8A	3A	4A	5A
Vitamin				
A, i.u.	2000	1960	1920	1880
D_3, i.u.	600	588	576	564
E, mg	25.0	24.5	24.0	23.5
Menaphthone, mg	1.3	1.27	1.25	1.23
Riboflavin, mg	4.0	3.92	3.84	3.76
Nicotinic acid, mg	28.0	27.45	26.88	26.32
Pantothenic acid, mg	10.0	9.8	9.6	9.4
Biotin, mg	0.05	0.049	0.048	0.047
Mineral				
Cu, mg	3.5	3.43	3.36	3.29
I, mg	0.4	0.392	0.384	0.376
Fe, mg	80.0	78.4	76.8	75.2
Mg, mg	300.0	294.0	288.0	282.0
Mn, mg	100.0	98.0	96.0	94.0
Zn, mg	50.0	49.0	48.0	47.0

Table 2.14. Performance of birds on Sand diluted diets at 4 weeks

Diet		1	3	4	5	
Sand g/kg		0	20	40	60	
Mean body weight kg	male	1.065	1.086	1.105	1.076	NS
	female	1.004	1.000	0.994	1.034	
Mean food intake kg	male	1.627	1.652	1.755	1.769	NS
	female	1.549	1.551	1.565	1.637	
Adjusted mean intake kg	male	1.627	1.619	1.685	1.663	NS
	female	1.549	1.520	1.502	1.538	
Mean fce.	male	0.65	0.66	0.63	0.61	NS
kg wt/kg intake	female	0.65	0.64	0.64	0.63	
Adjusted mean fce.	male	0.65	0.67	0.66	0.65	NS
kg wt/kg adjusted intake	female	0.65	0.66	0.66	0.67	

NS = not significant

Table 2.15. Performance of birds on Sand diluted diets at 8 weeks

Diet		1	3	4	5	
Sand g/kg		0	20	40	60	
Mean body weight kg	male	2.581	2.629	2.716	2.673	*
	female	2.198	2.245	2.224	2.276	
Mean food intake kg	male	5.933	6.319	6.406	6.650	**
	female	5.290	5.493	5.494	5.795	
Adjusted mean intake kg	male	5.933	6.193	6.150	6.251	**
	female	5.290	5.383	5.274	5.447	
Mean fce.	male	0.44	0.42	0.42	0.40	NS
kg wt/kg intake	female	0.42	0.41	0.40	0.39	
Adjusted mean fce.	male	0.44	0.43	0.44	0.43	NS
kg wt/kg adjusted intake	female	0.42	0.42	0.42	0.42	

NS = not significant

* = $p>0.025$

** = $p>0.01$

Table 2.16. Performance of birds on Energy and Protein reduced diets at 4 weeks

Diet		2	6	7	8	
%age Energy + Protein		100	98	96	94	
Mean body weights kg	male	1.061	1.103	1.062	1.025	**
	female	1.017	1.024	0.996	0.946	
Mean food intakes kg	male	1.616	1.665	1.610	1.653	NS
	female	1.500	1.507	1.523	1.445	
Mean fce. kg wt/kg intake	male	0.66	0.66	0.66	0.62	NS
	female	0.68	0.65	0.65	0.65	

NS = not significant

** = $p>0.01$

Table 2.17. Performance of birds on Energy and Protein reduced diets at 8 weeks

Diet		2	6	7	8	
%age Energy + Protein		100	98	96	94	
Mean body weights kg	male	2.554	2.588	2.582	2.467	***
	female	2.216	2.218	2.162	2.082	
Mean food intakes kg	male	5.965	6.171	6.044	5.947	**
	female	5.347	5.429	5.428	5.141	
Mean fce.	male	0.43	0.42	0.43	0.41	NS
kg wt/kg intake	female	0.41	0.41	0.40	0.41	

NS = not significant

** = $p>0.01$

*** = $p>0.005$

of the means may be estimated from the mean squares (MS) from the ANOVA tables (see Appendix) since the standard errors that are produced in the logarithmic scale are not useful when translated back into the original scale.

One set of control data was assigned to each of the sand-diluted diets and the ME and CP-reduced diets, giving data for 0 g sand/kg, and for 100% ME and CP.

Performance at 4 weeks (Tables 2.14 + 2.16)

Four week body weights were linearly depressed by the reduction of energy and crude protein. There were, however, no significant differences in 4 week body weights of birds fed sand-diluted diets. Neither energy and protein reduction nor sand-dilution had any statistical effect on mean food intakes over the four weeks. Feed conversion efficiency (fce) was reduced by the ME and CP-reduction but was unaffected by sand-diluted diets; as was the adjusted feed intakes and adjusted fce.

Performance at 8 weeks (Tables 2.15 + 2.17)

Whereas the reduction in ME and CP depressed eight week body weights curvilinearly, sand-dilution linearly increased eight week body weights. Correspondingly food intakes over the eight weeks had a curved response to ME and CP-reduction and was linearly raised by sand-dilution. Fce over the 8 weeks was reduced by the reduction in ME and CP but was unaffected by sand-dilution. Adjusted food intakes were raised by sand-dilution but there were no statistical differences in the adjusted fce of the sand-diluted diets and that of the two control diets.

TME values

The TME_N values of the diets are presented in Table 2.18. The values are corrected to zero nitrogen retention and are in terms of MJ/kg dry matter (DM). The TME_N values of the sand-diluted diets were calculated using adjusted feed intakes (i.e. total intake less diluent), giving the TME_N of the basal portion of the diet. The EEL_N values used in the calculations of TME_N for the starter and finisher diets were 71.50 and 66.38 kJ/48 hours respectively.

Table 2.18. TME_N and Crude protein values of the experimental diets

Starter diets 0-28 days

Diet	1	2	3	4	5	6	7	8
	Controls		Sand-diluted			ME, CP reduced		
TME_N MJ/kg DM	15.97		16.20	16.18	16.12	15.82	15.80	15.03
±SE	.13		.05	.10	.06	.05	.11	.10
Crude protein %	21.50		20.80	20.40	20.30	20.40	20.20	19.80

Finisher diets 28-56 days

Diet	1A	2A	3A	4A	5A	6A	7A	8A
	Controls		Sand-diluted			ME, CP reduced		
TME_N MJ/kg DM	14.87		14.86	15.26	14.79	14.28	14.65	14.43
±SE	.08		.15	.21	.14	.07	.16	.16
Crude protein %	19.00		19.50	18.80	18.20	18.90	18.20	17.80

For both the starter and finisher diets there were no statistically significant differences between the TME_N of the sand-diluted diets compared to the control diet, but the TME_N values of the ME and CP-reduced diets were significantly reduced ($p<0.01$, 0.05, respectively for the starter and finisher diets), as would be expected.

Mortality

Overall mortality during the eight weeks was 10.73%. There appeared to be an effect of dilution but, as mortality within the pens of control birds (diets 1 and 2) ranged from 5.42 to 14.58% and all other mortalities were within this range, the effect was considered to be a result of chance. Mortality during the eighth week alone was 4.22%, and this was judged to be the result of heat stress, and so mortality during seven weeks only was considered. This was 6.51% and there were no treatment effects. Mortality due to heat stress during the eighth week appeared to be concentrated at certain areas in the poultry house.

Discussion

Although it has long been considered that poultry utilise energy more efficiently from low energy-dense diets (Hill & Dansky, 1954), the results from this experiment strongly suggest that this is not the reason why sand-diluted diets improve broiler performance. Furthermore, the performance of birds fed energy and protein-reduced diets contradicts this hypothesis.

In this experiment sand-dilution significantly improved body weights at 8 weeks, whereas diets that were calculated to have the same ME and crude protein as the sand-diluted diets significantly depressed 8 week body weights.

The response of food intakes to diets reduced in ME and CP content was curvilinear, increasing at low levels of reduction but decreasing at the greatest reduction. The increase observed at low levels of reduction can probably be explained by the birds increasing their intake to satisfy their energy requirements. This effect could well have been expected to continue at the higher degree

of reduction. However, mean body weights of these birds were already significantly reduced at 4 weeks although the intakes up to this time had not been reduced significantly. The reduction in intakes by 8 weeks of age may have been a consequence of the smaller body weights of the birds and this would have perpetuated the depression in body weight.

Birds on sand-diluted diets were significantly heavier at 8 weeks than birds fed the control diet. Intakes over the eight week period were also significantly increased, but fce was unaffected. Adjusted (i.e. total intake less diluent) intake was, therefore, increased. Since fce was unaffected the increase in body weight was most likely due to the increased intakes, rather than a more efficient utilisation of the diet. Whether the presence of sand stimulated growth which in turn stimulated greater intakes, or whether the increase in intakes stimulated greater growth, is difficult to decide.

The TME values of the diets cannot be directly compared to the calculated ME values. Although TME and AME are related, according to the equation

$$TME = AME + EEL/FI$$

the ME values were calculated from figures derived by Bolton (1974) which were themselves calculated from the digestible protein, oil and carbohydrate of the feedstuffs and are not, therefore, AME values *per se*. Crude protein values also differed from the calculated values since mean values stated by Bolton were used in the calculation and the actual feedstuffs used naturally varied.

EXPERIMENT 2.8.

Objective

Experience at the PRC has shown that, if starved control birds are given glucose during a TME rapid bioassay, there is less individual variation in the EEL values. Furthermore, the birds suffer less stress measured in terms of weight loss. Experiment 2.8 was designed to investigate the effect of different glucose regimes during the starvation period upon EEL. Intact and caecectomised birds were used and the effect of caecectomy on endogenous energy loss was also determined.

Experimental

Design

Four treatments were randomly assigned to two groups of ten birds on two separate occasions. The treatments were:-

1. 25 ml glucose solution (500 g/kg) was tube-fed twice during the 48 hour starvation period, at about 6 and 30 hours, followed by 40 g glucose at the start of the 48 hour collection period.
2. 25 ml glucose solution (500 g/kg) was tube-fed twice during the 48 hour starvation period, at about 6 and 30 hours.
3. 25 ml water was tube-fed twice during the 48 hour starvation period, at about 6 and 30 hours, followed by 40 g glucose at the start of the 48 hour collection period.
4. 25 ml water was tube-fed twice during the 48 hour starvation period, at about 6 and 30 hours.

Birds and management

20 dubbed, adult White Leghorn cockerels, 10 intact and 10 caecectomised, were individually caged and starved for 48 hours. During this period, at about 6 and 30 hours after housing, the birds were twice tube-fed 25 ml glucose solution (500 g/kg) or 25 ml water, according to a previously randomised plan. After 48 hours the birds in groups 1 and 3 were tube-fed 40 g glucose. All birds were

returned to clean cages with a clean tray and the time of housing recorded. The excreta voided over the following 48 hours were collected quantitatively. The samples were freeze-dried, allowed to equilibrate to atmospheric moisture, weighed and then analysed for gross energy and total nitrogen.

The procedure was repeated two weeks later with each bird receiving a different treatment.

Results

The values for Endogenous Energy Loss (EEL) and nitrogen-corrected EEL (EEL_N) are presented in Tables 2.19 and 2.20 respectively.

The distribution of EEL is assymetrical, some birds having very high values. This may be due to some birds experiencing greater stress although kept under the same circumstances. It was, therefore, envisaged that glucose might have a greater sparing effect on these extreme birds. For this reason, for analytical purposes, the values of EEL and EEL_N were transposed into logarithms which had the effect of minimising the bird to bird variation without masking the effect of glucose. The fitted values presented in the tables are the mean values obtained from the logarithmic values.

The presence of glucose during the starvation period significantly reduced the EEL from both intact and caecectomised birds ($p<0.05$).

There was also a significant glucose x week interaction ($p<0.05$), the glucose treatment having a greater sparing effect during the second week of the experiment.

Birds which had been caecectomised had significantly greater energy losses over both experimental periods ($p<0.01$).

Correcting the EEL values to zero nitrogen retention (EEL_N) removed all of the above effects except that of caecectomy.

Discussion

It is generally considered (Dale & Fuller, 1981) that glucose is completely metabolised by poultry and makes no contribution to either their exogenous or endogenous energy losses. The results from

Table 2.19. Mean Endogenous Energy losses from Intact and Caecectomised birds on four glucose routines (kJ)

Treatment	1	2	3	4
Week 1				
Intact				
observed values	64.19 ±SE 9.08	63.29 ±SE 2.85	53.67 ±SE 2.71	75.86 ±SE .47
(ln)	(4.1517 ±SE .2014)	(4.1457 ±SE .0771)	(3.9815 ±SE .0713)	(4.3287 ± SE .0109)
fitted values	59.03	63.90	59.70	73.00
Caecectomised				
observed values	66.85 ±SE 4.29	78.90 ±SE 8.31	82.02 ±SE 13.34	85.62 ±SE 4.69
(ln)	(4.2003 ±SE .0909)	(4.3574 ± SE .1779)	(4.3936 ± SE .2321)	(4.4469 ± SE .0971)
fitted values	71.80	81.00	72.60	88.70
Week 2				
Intact				
observed values	60.41 ±SE 2.86	68.65 ±SE 9.02	79.77 ±SE .44	96.97 ±SE 23.04
(ln)	(4.0979 ±SE .0916)	(4.2203 ± SE .1867)	(4.3791 ± SE .0078)	(4.5454 ± SE .3426)
fitted values	59.30	64.50	76.60	108.00
Caecectomised				
observed values	70.67 ±SE 2.07	74.37 ±SE 3.31	91.48 ±SE 8.88	150.42 ±SE 4.70
	(4.2571 ± SE .0502)	(4.3081 ± SE .0630)	(4.5059 ± SE .1175)	(5.0129 ± SE .0442)
fitted values	72.10	78.40	93.00	131.20

Table 2.20. Mean Endogenous Energy losses from Intact and Caecectomised birds on four glucose routines corrected to zero N_2 retention kJ

Treatment	1	2	3	4
Week 1				
Intact	29.97 ±SE 6.5	29.82 ±SE 4.29	20.82 ±SE 2.19	26.54 ±SE 1.68
Caecectomised	31.60 ±SE 1.72	32.01 ±SE 2.63	40.24 ±SE 5.25	34.61 ±SE 3.53
Week 2				
Intact	28.42 ±SE .58	28.07 ±SE 8.32	33.00 ±SE 1.62	21.49 ±SE 1.90
Caecectomised	39.49 ±SE 2.67	30.70 ±SE 5.71	41.27 ±SE 5.57	28.92 ±SE 2.64

this experiment support this hypothesis. Indeed, birds given glucose during the starvation period lose less energy than birds which have received nothing at all. The energy supplied by the glucose presumably reduces the need for the birds to catabolise their body reserves to provide for their maintenance energy requirements. The disappearance of this glucose-sparing effect on EEL by correcting the figures to zero nitrogen balance (EEL_N) suggests that the differences observed in EEL between starved birds and those receiving glucose are caused by greater losses in nitrogen from the starved birds as a result of greater protein catabolism.

This raises the question as to whether EEL values, which are uncorrected for differences in nitrogen retention and are determined with starved birds (Sibbald, 1976, 1981; Tenesaca & Sell, 1981), accurately reflect the endogenous energy that is lost from birds given an energy source in the form of a feedstuff either by tube or under more normal feeding practice. Under normal ad lib. feeding, birds catabolise minimal amounts of body protein and it may be that the EEL measured from starved birds is artificially high. If it is also higher than that from birds fed small amounts of feeds through a tube, as in the TME assay, the effect would result in attributing higher TME values to the materials under test.

This criticism would be overcome if the data are corrected to zero nitrogen balance. From this experiment it appears that nitrogen correction removes the effect of starvation on EEL and effectively removes the value of the glucose treatments. However, the application of the glucose treatment would still be advisable because the birds, being under less stress, lose less body weight during the test period. This could be an important factor in maintaining the health of the test flock, especially if the birds are used frequently for such assays.

The sparing effect of glucose was greater during the second week of the experiment. The effect of treatment 1 over treatment 4 was to reduce the mean EEL from intact birds by 36.56 kJ in week 2 compared to 11.47 kJ in week 1. External temperatures during the second week were more extreme and the greater difference observed was most probably caused by the very low temperatures experienced that week increasing the birds' maintenance requirements and leading to an increase in protein catabolism. This effect of temperature

does confirm the findings of Dale & Fuller (1981) and emphasises the need for maintaining a starved control group of birds during each TME bioassay rather than applying a standard value to each set of data. Nitrogen correction, however, does minimise differences caused by temperature effects.

Caecectomy significantly increased the EEL and this effect was not removed by the correction to zero nitrogen retention. Caeca are responsible for a considerable amount of water reabsorption and consequently caecectomised birds lose a greater amount of water. Although the moisture content of the excreta samples was not determined the excreta from the caecectomised birds was visibly wetter than the excreta from the intact birds, not only in this experiment but also in Experiments 2.5 and 2.6. It is possible that the increase in water moving through the large intestine would increase the sloughing off of the epithelial cells lining the wall, thereby increasing the EEL. However, this would probably not contribute enough to increase substantially the EEL, especially when the shortness of the large intestine is considered. McNab (1973) concluded that the caeca may play a compensatory role when normal digestive processes are impaired, conserving water, nitrogen and energy. It is possible that the caeca of the intact birds undertake this function during the stress of the starvation period, thereby reducing the EEL. Again the difference between the intact and caecectomised birds is greater in the starved group (treatment 4) than in the treatment 1 birds that were considered to be under least stress.

DISCUSSION

Dietary Dilution

Sand

Reports concerning the beneficial use of sand in poultry diets have mainly related to increases in feed conversion efficiency and improved energy utilisation. In general, it has been found that sand can be added in low levels to poultry diets without having any adverse effects upon growth or laying performance, whilst actually decreasing the amount of feed or energy required for these purposes. Improved growth has only been reported by Rowland & Hooge (1980) who claimed that levels of 40 and 100g sand/kg improved chick growth compared to that of birds receiving the control diet or one containing 20g sand/kg. However, these differences were not significant. In fact, in the same paper the authors referred to a trial in which 60g sand/kg was found to depress body weight. A significant depression in growth was also reported by Sloan & Harms (1974); 100g sand/kg included in a low protein diet (200g CP/kg) significantly reduced 4 week body weight. Most published reports have, however, indicated that the presence of sand in poultry diets has no effect upon body weights. Dietary sand at varying levels of inclusion have been reported to have no significant effect upon the growth of broiler chicks (Andrews *et al.* 1972; Sellers *et al.*1979, 1980), broiler breeders (Voitle *et al.* 1974; Hogsette *et al.* 1976) or turkey poults (Voitle & Harms, 1976; Harms & Voitle, 1977; Miles *et al.* 1978, 1980). Experimental results given in this thesis agree with the maintained growth of chicks receiving sand-diluted diets claimed by the above workers, at least in the early stages of growth, up to 4 weeks of age. In Experiment 2.7, the four week body weights of broilers receiving diets containing sand in levels of 20, 40 and 60g/kg were numerically, but not statistically significantly, higher

than those of the control birds. At eight weeks, however, the same birds had body weights that were significantly higher than those of the control birds (p 0.025). This increase was, however, accompanied by an increase in basal feed intakes and the feed conversion efficiencies were not significantly different from that of the control birds.

The increased feed conversion efficiencies normally referred to in work involving dietary dilution by sand have not been confirmed by studies reported in this thesis. In two experiments (2.1 and 2.7) there were no significant differences between the basal fce's of the sand-diluted diets and the fce of the control diet. Improved fce and improved energy utilisation due to the presence of sand in poultry diets have been quoted for growing chicks (Andrews et al.1972; Hooge & Rowland, 1978; Harms & Damron, 1973) laying hens (Harms & Damron, 1973; Hooge et al. 1977; Hooge & Rowland, 1978) and turkey poults (Harms & Voitle, 1977). Such improved efficiences of feed and energy utilisation indicate that the basal feed intakes of birds receiving sand-diluted diets are maintained, or even reduced. Hooge (1979) quotes unpublished results of Harms & Damron (1974) in which dietary sand at a level of 25g/kg reduced actual feed consumption (i.e. feed minus diluent). A decrease in actual feed consumption was also reported by Hooge & Rowland (1978) and Hooge et al. (1979). These incidences of decreased actual (basal) feed consumption all occured in experiments with laying hens and not growing chicks. Studies reported in this thesis have indicated a statistically significant increase in basal feed intakes by birds receiving sand-diluted diets (Experiment 2.7). These results dispute the claim by Hooge (1979) that sand would be economical to use as a dietary diluent despite numerically lower weight gains since less actual feed would be required for a bird to reach a given weight.

Results in this thesis regarding the metabolisable energy value of sand-diluted diets are ambiguous. The AME value of a diet has been found to be maintained by

sand-dilution, as too have the TME values of three diets (Experiments 2.2 and 2.7) determined by the traditional Sibbald method. However, the estimated TME of sand-diluted diets in Experiment 2.1 was greater than that of the control, but not significantly so, and the TME of sand-diluted diets obtained by linear regression of energy balance at three levels of intake (Experiment 2.5) was statistically significantly greater than that of the control diet. Sibbald (1980) found no evidence that sand influenced the TME of various grains. Miles et al. (1981) reported that the inclusion of 25g sand/kg in a diet numerically improved the TME but this was not statistically significant. Evidence is presented in this thesis showing that sand has no effect on endogenous energy loss (table 2.5) and this is confirmed by Sibbald (1980). The experiments which indicated that sand improves dietary TME were carried out over a period of time, unlike the normal TME assay. It is possible that, under more normal conditions of feeding, sand aids digestion thereby increasing the TME. Miles et al. (1981) suggested that sand might improve grinding of food in the gizzard thereby creating a larger surface area for the food particles, enabling a greater contact with the digestive juices. It has also been suggested that the fineness of the particles of sand contribute to its mixing with the food particles, again increasing the surface area available for digestion. It has been reported that the improvement in feed and energy utilisation increases with increasing fineness of sand (Oluyemi & Harms, 1977). If, however, TME is increased by the presence of sand, even with maintained basal intakes, it could be expected that growth would also increase. Results presented in this thesis only show increased growth with increased intakes and a maintained TME (table 2.15). It can probably be concluded, therefore, that the presence of sand in a typical poultry diet does not have any effect upon dietary TME.

There is no evidence relating to the action of sand in improving feed efficiency and/or growth. Cooley & Burroughs (1962) found that the addition of sand to ruminant diets increased body weights and feed utilisation but later it was found that the major increase in weight was due to the retention of sand in the rumen. There is no evidence of sand being retained to this extent in the intestine of the chicken. Oluyemi et al. (1978) reported that sand did not accumulate to any substantial extent in the gizzard or any other part of the digestive tract and could not account for any increase in body weight. Many workers (Harms et al. 1974; Damron & Harms, 1976; Damron et al. 1976; Hooge, 1979) suggest that sand acts by diluting the energy density of the diet and quote Hill & Danskys' work (1954) demonstrating that poultry utilise energy more efficiently from low energy-dense diets. However, the results reported in this thesis strongly refutes this. In Experiment 2.7, when the action of diets containing 20, 40 and 60g sand/kg was compared to that of diets containing calculated equivalent amounts of ME and crude protein, the results were completely the opposite. Energy- and protein-reduced diets had a curvilinear effect on food intakes and significantly depressed 8 week body weights, whereas the sand-diluted diets significantly increased body weights and food intakes. However, it is not necessarily correct to compare the sand-dilution work with that of Hill & Dansky (1954). These authors reduced the energy density of their diet by including large quantities of pulverised oats, obtaining optimum growth when the diet was diluted by 300 and 400g oats/kg, which are far higher levels of inclusion than normally used in studies on dietary dilution by sand.

It seems reasonable to conclude that sand can be included in a typical broiler diet at up to a level of 60g/kg without detrimental effects, although 40 g sand/kg would appear to be optimal in terms of improved growth. Feed conversion efficiency is not, however,

improved. The increase in 8 week body weights in Experiment 2.7 was most likely a result of the increased basal intakes, since fce was maintained. However, it is not obvious whether the increase in weight preceded increased intakes or vice versa. Body weights of the sand fed birds at 4 weeks were not statistically significantly different from those of the controls although there did tend to be a numerical increase in the former. This would probably have stimulated an increase in intakes which in turn would have resulted in improved growth.

Whether or not sand would prove to be economical to add to broiler diets would depend on feed prices and the wholesale selling price of the birds prevalent at the time. Since the increase in weight gains of broilers in Experiment 2.7 occured during the last four weeks of production, it may be possible to obtain birds ready for market at, perhaps, seven weeks, thereby saving one weeks supply of food. The possibility of this may be determined by weekly weight measurements during an 8 week trial involving sand-diluted broiler diets.

Kaolin

Results of studies concerning the effects of the addition of kaolin to poultry diets vary considerably. Ousterhout (1970) has quoted results ranging from no effect to improved performance and has suggested that the average result shows that 50g kaolin/kg decreases feed conversion efficiency by about 2% whilst having no significant effect on growth. Previously Ousterhout (1969) had fed diets containing between 5 and 400g kaolin/kg to broiler chicks and reported that efficiency was improved up to the 160g/kg level. Improved performance has tended to have been observed in terms of feed efficiency or energy utilisation. Matterson et al. (1972) reported that the weights gained by broilers fed a diet containing 60g kaolin/kg from 0 to 4 weeks of

age were not significantly different from those gained by control birds but that the fce was improved by about 5%. Spandorf et al. (1972) found that kaolin significantly improved feed utilisation by laying hens and Harms & Damron (1973) reported that the addition of 25g kaolin/kg to the diet of laying hens improved energy utilisation by about 5%.

In the experiment reported in this thesis the inclusion of kaolin in levels ranging from 20 to 60g/kg in a practicle broiler starter diet significantly reduced body weight gains at 21 days of age. Day et al. (1970) showed that chick growth was depressed after feeding diets containing 50 to 100g kaolin/kg, although at levels of 10 and 20g/kg performance was improved. Alquist et al.(1967) had reported that optimal performance was obtained by feeding a diet containing 20g kaolin/kg. The growth depression reported in this thesis, however, was evident at the 20g/kg inclusion level and so the results were not a consequence of excessively high levels of kaolin being used. Indeed, Spandorf (1973) found that the growth of birds receiving 60 g kaolin/kg was equal to that of control birds and Charles & Widley (1975) reported that neither 25 or 50g kaolin/kg in a layer diet had any significant effect upon egg production, fce or body weight.

Ousterhout (1970) has claimed that the inclusion at a level of 50g/kg of a number of commercially available kaolins resulted in a range of performances including a dilution effect on fce coupled with a slight decrease in body weight. The results of Experiment 2.1 however showed a depression in weight gain greater than that anticipated from simple dilution. Expected values for weight gains would have been 379, 372 and 364g for birds fed diets containing 20, 40 and 60g kaolin/kg respectively, whereas the observed values were 356, 362 and 330g respectively. Basal food intakes were significantly less than those of the control birds. It can be seen from the results that the growth

depression is especially severe at the 60g/kg inclusion level and corresponds with the greatest reduction in basal food intakes. It is possible that since the AME values are not significantly different to that of the control diet, the severe depression of growth at the 60g/kg inclusion level results only from the very low feed intake. It is possible that the bulky nature of the kaolin contributes to the results by preventing the birds from consuming sufficient food to meet their energy requirements.

Published data relating to the use of kaolin in poultry diets have tended to be contradictory and the results presented here do not help to resolve this. However, in view of the contradictory nature of the published work, the growth depressing action of kaolin demonstrated here would suggest that it has little, if any, value in poultry diets.

Cement Kiln Precipitator Dust

Reports concerning the growth stimulating action of cement kiln dust have not been consistent, either between species or within them. Improved growth due to the inclusion of CKD in the diets of ruminants has been demonstrated in both steers and sheep (Wheeler & Oltjen, 1978; Roginski & Wheeler, 1978) but in the case of monogastric animals, including both rats and chicks, the results have been contradictory. Thus,Roginski & Wheeler (1978) demonstrated a significant improvement in growth when rats were fed a purified diet supplemented with 10g CKD/kg, whereas Gold et al.(1979) failed to show any improvement in performance when CKD was included in a practical diet fed to adult female rats. Veltmann & Jensen (1980) found no evidence of growth promoting properties of CKD in trials with broiler chicks fed practical diets. These authors, on evaluating varying levels and different sources of CKD, found that 30g/kg or more of CKD in a practical diet unbalanced

in calcium and phosphorus resulted in depressed growth and produced a rachitic condition. However, with a diet balanced in calcium and phosphorus both 15 and 30g CKD/kg had no effect on chick performance between 3 and 4 weeks of age. In contrast, however, Kienholz (1978) reported significant weight gains and improved feed efficiency in chicks fed a diet containing 15g CKD/kg.

Results reported in this thesis (table 2.2) tend to agree with the growth depression observed by Veltmann & Jensen (1980). Chicks fed CKD-diluted diets from 1 to 3 weeks of age were significantly lighter than birds fed control diets. The fce values of birds receiving CKD-diluted diets were signifiantly lower than those of birds receiving the control diet. Since both the AME and estimated TME values of the control diet were not significantly different from those of the other treatments, including CKD, this would indicate a less efficient utilisation of energy by birds receiving CKD-diluted diets. This may have been caused by an increased loss of energy from the body - diverting energy away from growth purposes. Feeding activity of the birds receiving CKD-diluted diets was somewhat frenetic; spillage was considerably greater than that among other groups of birds. It is possible that this feeding behaviour may have increased maintenance requirements resulting in less energy being available for growth.

The potential toxic nature of CKD cannot, however, be ruled out as a cause of the growth depression. Apart from the increased feeding activity birds receiving CKD were less active than other birds. However, there were no mortalities over the fourteen day experimental period, but the birds behaviour was adjudged to be subdued, indicating that some other physiological effect was occuring in addition to growth depression. With hindsight a mineral analysis of the CKD might have pointed to the presence of toxic levels of certain elements. It is interesting to note that the growth depressing action of CKD in the studies of Veltmann &

Jensen (1980) only occured when the diets were unbalanced in calcium and phosphorus. The Ca:P ratio was considered to be balanced in the diets used in this study and it seems unlikely that adverse Ca:P concentrations have contributed to the poor performance observed. The composition of dusts from different sources have been shown to vary considerably (Wheeler & Oltjen 1978) and this may be the cause of conflicting results obtained by different workers. It may be that only some sources of CKD exhibit growth enhancing properties. It is important that in any future work involving CKD that each sample should be analysed for any toxic levels of minerals and careful attention paid to the dietary calcium:phosphorus ratios.

Cellulose

Although results presented in this thesis (table 2.2) show that the inclusion of cellulose in a practical broiler diet significantly depresses body weight gain, there have been other conflicting reports. Sibbald et al. (1960) reported a small depression in body weight gains when cellulose was included at a level of 60g/kg in the diet of growing chicks. This growth depressing action became increasingly severe at levels of 120g/kg and more. Andrews et al. (1972) also reported a depression in growth when cellulose was used as an inert filler and a marked reduction in growth was reported by Bayer et al. (1978) when 60g cellulose/kg was included in a chick starter diet at the expense of maize. Davis & Briggs (1948) however, demonstrated growth depression only when levels of 200g cellulose/kg were included in the purified diet of growing chicks. Cellulose at levels of between 50 and 150g/kg actually caused a significant increase in growth and feed efficiency. This beneficial action may, however, have resulted from the presence of fibre in an otherwise fibre-free diet rather than from the action of cellulose per se. A growth stimulating

effect of cellulose has also been recorded by Saito et al. (1959); 35, 95, 165 and 265g cellulose/kg improved growth above that of birds receiving a control diet containing no cellulose, but there were no significant differences between the cellulose treatments. There have also been, however, reports that cellulose has neither a depressing nor a stimulating effect on chick growth (Begin, 1961; Yoshida & Hoshii, 1970; Akiba & Matsumoto, 1978).

A reduction in the utilisation of the diet, indicated by growth depression coupled with the maintenance of basal feed intakes and the metabolisable energy value of the diet, has been demonstrated by Dvorak & Bray (1978). When cellulose was included in a chick starter diet at levels of 100, 200 and 300g/kg, basal feed intakes were maintained but growth was depressed, indicating a reduction in utilisation. Dvorak & Bray (1978) suggested that the reduced utilisation was in fact a reduction in the absorption of the diet, perhaps due to increased transit time or a physical action within the gut. However, studies reported in this thesis demonstrate that cellulose does not affect the metabolisable energy value of the diet. Three out of four experiments showed a maintenance of either AME or TME. This is confirmed by studies of both Sibbald (1980) and Miles et al. (1981). Sibbald (1980) included 0 to 4 g of cellulose in 25g of a layers' diet or maize and found that the TME of neither was changed by the presence of cellulose, concluding that cellulose has no available energy nor does it affect the availability of dietary energy. Miles et al. (1981) combined the TME calculation of Sibbald (1976) with the meal-feeding technique of Farrell (1978) to determine the TME of a turkey poult diet containing 25g cellulose/kg. The TME was reduced but not significantly so. That cellulose has no available energy for poultry was also demonstrated in an experiment reported in this thesis involving a TME determination on intakes of 10, 20 and 30 g of cellulose (table 2.11). There were no statistically significant

difference in the endogenous energy losses of birds receiving the three input levels in the case of intact birds. There was an increase in the EEL of caecectomised birds with increasing inputs of cellulose; this may have been caused by the bulking action of cellulose increasing gut transit time and/or incresing exfoliation of the gut mucosa. It may have been, however, due to the effect of caecectomy rather than the actual cellulose. The lack of effect of cellulose upon EEL was also confirmed by Sibbald (1980). Potter et al.(1960) suggested that cellulose possessed a negative ME value, -47 ± 21J/g, and proposed that this was due to cellulose absorbing nutrients and making them unavailable to the bird. In light of more recent knowledge, however, it is more likely that this figure represents the endogenous energy loss. Although there was a slight numerical increase in the dietary TME at the 60g/kg inclusion level of cellulose (Experiment 2.2) it is unlikely that there is any digestion of cellulose by poultry. Despite a numerical increase in the length of caeca of birds receiving a cellulose-diluted diet, in trials involving caecectomised birds there was no evidence of cellulose improving the ME of a basal diet nor of any digestion of cellulose itself within the caeca.

There is little evidence to indicate the means by which cellulose depresses growth. In Experiment 2.1, the growth depression was greatest at the 60g/kg level , which corresponded to the greatest depression in basal intakes. It is possible that birds receiving 60g cellulose/kg were unable to increase their food consumption sufficiently to maintain their energy intakes and that this was the cause of the growth depression. Indeed, over the two week experimental period, birds receiving the control diet consumed 8.78 MJ whilst those receiving the diet containing 60g cellulose/kg consumed 8.45 MJ. Sibbald et al. (1960) found that the volume of feed consumed by growing chicks was highly correlated with body weight and suggested that birds were initially

unable to increase the volume of intake sufficiently and this led to depressed growth, which in turn led to lower intakes perpetuating the growth depression. This is probably not the whole explanation however, since together with the reduction in energy intakes there is also a reduction in the efficiency of energy utilisation. Birds receiving no cellulose gained 44g per MJ intake, whereas those receiving 60g cellulose/kg gained only 38g/MJ. Since there was a numerical increase in the size of the caeca due to the presence of cellulose it is possible to assume that this could also have occured in other parts of the gut. It has been suggested that the water absorbing property of cellulose and other fibrous substances causes an increase in the size of the gut (Savory & Gentle, 1976a; Hedge et al. 1978; Dvorak & Bray, 1978). This could increase the birds energy requirements for maintenance and could perhaps account for, at least in part, the apparent reduction in energy utilisation.

Sawdust

It has generally been considered that high levels of fibre in poultry diets are detrimental to growth, feed conversion efficiency and egg production (Heuser et al. 1945; Scott et al. 1947; Vlcck, 1970; and El-Kotoury et al. 1973) but there have been reports that the addition of sawdust to poultry diets has either a beneficial effect on growth or, at least, no effect at all. Ewing (1947) reported that spruce wood flour at a level of 202 g/kg had no deleterious effects on chick growth and actually resulted in a slight increase in growth at 28 weeks. Davis & Briggs (1948) also reported that the inclusion of sawdust in a purified diet up to a level of 200g/kg had no adverse effects. Lower levels of sawdust have also been claimed to be beneficial. El-Abbady et al.(1973) reported that both levels of 40 and 105g sawdust/kg improved body weights whereas 200g sawdust/kg depressed body weights of Baladi

Whites at 20 weeks. Sawdust at a level of 60g/kg was found to maintain body weights of White Austrian Turkeys from 4 to 24 weeks of age when included in isoenergetic, isonitrogenous diets.(Omar et al. 1973) although higher levels depressed growth. Likewise, 60g sawdust/kg in a general layers' diet had no effect on body weights but increased feed conversion efficiency (Deaton et al. 1976).

Results presented in this thesis (table 2.2) do not confirm the beneficial action of sawdust with birds of a fast growing broiler strain. Indeed, three week body weights were significantly depressed when sawdust was included in a typical broiler syarter diet. Basal food intakes were also significantly lower than those of birds receiving the control diet. Although the fce of the birds receiving the control diet was not significantly greater than the treatment means of the diluted diets in Experiment 2.1, the fce values of birds receiving sawdust-diluted diets are lower than the control values which suggests some degree of reduced efficiency of energy utilisation since AME and $\hat{TME}$ values are not significantly different. Indeed, the numerically higher $\hat{TME}$ values would further indicate this. It is possible that the nature of the sawdust would prevent the birds from consuming sufficient feed to meet their energy requirements or may even increase them. Maintenance requirements may have been increased because of an increase in digestive activity, such as prolonged grinding in the gizzard, due to the nature of sawdust. It is also possible that there is an energy cost associated with simply moving the sawdust down the digestive tract. Deaton et al.(1976) reported an increase in gizzard weights in layers fed a diet containing 60g pine shavings/kg. It is also possible that the bulk of a sawdust containing diet would increase the dimensions of other parts of the gut (Hedge et al. 1977). Enlargement of the alimentary canal could increase energy requirements for maintenance, thus diverting energy away from growth,

although it is not possible to say whether this could account for the size of reduction in growth reported for the 60g/kg inclusion level of sawdust. It might be possible to ascertain whether or not the presence of sawdust in a broiler starter diet increases the birds' energy expenditure by using a method of direct calorimetry. In the context of this study, however, it can be concluded that sawdust is not suitable for use as a diluent in poultry diets.

The role of the avian caeca has been a controversial subject for many years, but generally the caeca are considered to be of little nutritional importance since performance is apparently unaffected by their removal. A number of workers have, however, indicated that the caeca are responsible for the digestion of at least some of the crude fibre in the diet (Mangold, 1934; Halnan, 1949; Thornburn & Willcox, 1965a). There is some evidence that the size of the caeca can be influenced by the diet, especially by the crude fibre content, and that this can affect their function (Lewin, 1963; Pendergast & Boag, 1973; Savory & Gentle, 1976a). However, the results of experiments described in this thesis did not confirm this. Although both sand- and cellulose-diluted diets were associated with a numerical increase in the length of the caeca of three week old chicks above that of control birds, this increase was not statistically significant and therefore inconclusive.

Thornburn & Wilcox (1965a) reported that caecectomy reduced whole diet digestibility, including the digestibility of crude fibre. The digestibility of crude fibre was apparently dependent upon the nature of the food and its crude fibre content. This would confirm reports of studies by Radeff, cited by Mangold (1934) which demonstrated that neither intact nor caecectomised birds digested the crude fibre of barley, whereas the digestibility of the crude fibre of wheat was greatly reduced after caecectomy. However, in a study reported in this thesis, involving the use of intact and caecectomised cockerels to determine the TME of a conventional diet and diets diluted by either sand or cellulose, there was no evidence of any effect of caecectomy. Thornburn & Willcox (1965a) reported that cellulose digestibility was reduced in individual birds after caecectomy but that this was not always evident when results were compared to those of intact birds. However, the results of Experiment 2.6 in this thesis indicated that cellulose was not digested by either caecectomised or intact birds. However, individual birds were not studied before caecectomy and so the results of Thornburn & Willcox cannot be disputed. It should be pointed out, however, that Thornburn & Willcoxs' work involved the determination of the cellulose component of the diet and subsequent droppings, whereas the studies reported

here involved the use of pure cellulose. The lack of effect of caecectomy upon crude fibre digestibility has also been demonstrated by Nakahiro et al. (1974) and Nakahiro & Isshiki (1975).

It is generally concluded that the caeca play an important role in water reabsorption, both of intestinal and urinary origin (McNab, 1973). Olson & Mann (1935) quote reports by Röseler (1929) demonstrating that caecectomy considerably reduces the percentage of dry matter in the faeces compared to that of intact birds. This reduction in faecal dry matter was also reported by Thornburn & Willcox (1965a). Although the dry matter of droppings were not recorded in the studies undertaken here with caecectomised birds, the increased moisture content of the droppings from caecectomised birds was visibly obvious.

McNab (1973) concluded that although the caeca are unnecessary for the optimal growth of domestic birds they may have a definite role in the wild state, enabling the conservation of water, nitrogen and energy, and may also play a compensatory nutritional role when normal digestion is impaired. Support for this conclusion has been shown by work presented in this thesis concerning the use of glucose during the TME bioassay (Experiment 2.8). Both intact and caecectomised birds were used in this study and it was found that, regardless of the presence or absence of glucose, caecectomised birds had significantly greater energy losses than intact birds ($p<0.01$). In view of the fact that the birds are under considerable stress during a TME assay, because of the severe restriction on energy intake, it is quite possible that the caeca of the intact birds perform some compensatory role by retaining energy. Thus the intact birds are excreting less energy, rather than the caecectomised birds excreting more.

3. STUDIES ON GUAR GUM

EXPERIMENT 3.1.

Objective

A dose response trial was carried out in order to determine the effect of different concentrations of guar gum upon chick growth and on the AME value of a typical broiler starter diet. The effect of guar gum upon nitrogen retention and fat digestibility was also determined.

Experimental

Design

Eight treatments were each randomly assigned to two groups of three, single-sexed, birds in each of three blocks, giving six replicates of each treatment. The treatments were:-

1. Basal diet (Control)
2. 2.5g guar gum/kg basal diet
3. 5.0g " "
4. 7.5g " "
5. 10.0g " "
6. 12.5g " "
7. 15.0g " "
8. 17.5g " "

Birds and management

About 200 day-old Marshall stock, male broiler chicks from the same hatch were wing-banded and reared until 7 days of age in thermostatically controlled battery brooders. At 7 days of age the chicks were individually weighed; 144 chicks from the middle weight band were selected and distributed to one of 24 double cages, such that there were three birds per half cage, according to a previously randomised plan. The cages were the same as those described in Experiment 2.1.

Food and drinking water were available ad lib at all times. Total food intake per group of 3 birds was recorded over a 14 day period,

from 7 to 21 days of age. The birds were individually weighed at the end of the 14 day period. An **AME assay** was carried out during the last four days of the experiment. The wire floors of the cages were cleaned and a clean tray placed underneath. All excreta from each group of six birds were collected quantitatively at the end of the four day period, and total intake per group of six birds was recorded.

The excreta were oven-dried, allowed to equilibrate to atmospheric moisture and analysed for gross energy and total nitrogen for the determination of AME_N and nitrogen retention. The samples were also analysed for fat content for the determination of fat digestibility.

Diets

The control diet was formulated to the same composition as the diet used in Experiment 2.1 (Table 2.1). The seven test diets consisted of the control diet containing 2.5 to 17.5 g/kg of powdered guar gum, in 2.5 g/kg increments.

The diets were presented to the birds in mash form. All diets were analysed for dry matter, gross energy, total nitrogen and fat content.

Results

Mean body weight gains over the two week experimental period are presented in Table 3.1. There was a significant ($p<0.01$) depression in body weight gains but this was not accompanied by a depression in food intakes.

The results of the **AME assay** are given in Table 3.2. The AME is presented as MJ/kgDM and is corrected to zero nitrogen retention (AME_N). Nitrogen retention and fat digestibility are also presented on a dry matter basis. Linear regression indicates a significant depression in AME_N, nitrogen retention and fat digestibility ($p<0.001$, 0.01, 0.001, respectively). There are no significant differences in the food intake figures.

Table 3.1. Mean bodyweight gains and mean food intakes over 14 days

Diet		1	2	3	4	5	6	7	8	
Guar gum	g/kg	0	2.5	5.0	7.5	10.0	12.5	15.0	17.5	
Weight gain	g	382	403	387	359	325	286	289	260	**
	±SE	15	14	14	15	14	14	14	14	
Food intake	g	534	568	528	489	514	470	503	474	NS
	±SE	29	27	27	29	27	27	27	27	

NS = not significant

** = $p < 0.01$

Table 3.2. AME_N, Nitrogen retention and fat digestibility of diets containing Guar gum

Diet	1	2	3	4	5	6	7	8		
Guar gum g/kg	0	2.5	5.0	7.5	10.0	12.5	15.0	17.5		
Intake over AME assay, g	887	1017	1004	909	868	822	894	938	±SE 103	NS
AME_N MJ/kgDM	12.98	13.04	12.87	12.73	12.57	12.36	12.19	11.92	±SE .16	***
Nitrogen retention g/kg food intake DM	23.34	22.91	23.79	22.78	20.84	22.61	21.71	18.47	±SE .99	**
Fat digestibility g/kg food intake DM	99.49	109.33	97.72	92.74	82.54	89.98	85.24	77.28	±SE2.79	***

NS = not significant

** = $p < 0.01$

*** = $p < 0.001$

Discussion

The depression in body weight gain under conditions of maintained food intakes suggest a depression in either energy intake or energy metabolism due to the presence of guar gum. The results here indicate that the energy metabolism is reduced since there is a severe depression in AME.

Neither nitrogen retention nor fat digestibility show a clear pattern of change despite both being significantly depressed. Univariate analysis of variance suggests that the AME_N was only affected by the presence of guar gum whereas both nitrogen retention and fat digestibility were influenced by their respective dietary inputs, increasing with increased inputs. Thus there was numerical masking of the results.

Both nitrogen retention and fat digestibility can be expressed in terms of energy, i.e. MJ/kg and the range of values then expressed as a percentage of the range in AME_N:- N_2 retention = 8.6%
Fat digestibility = 65.0%.
Thus it appears that the major effect of guar gum is to depress fat digestibility. This can be confirmed by a statistical analysis involving the removal of each parameter in turn from an analytical model comparing AME_N with the dietary inputs and nitrogen retention and fat digestibility. Only the removal of the fat digestibility parameter gives a statistically significantly poorer fit than the original model ($p<0.05$).

It appears, therefore, that guar gum reduces the absorption of the whole diet, mainly affecting the fat digestibility, leading to a reduction in AME_N and a reduction in body weight gain. Since absorption of the whole diet is impaired it would not appear to be a localised action. An action involving the suppression of digestive enzymes is also unlikely since it would not only involve all groups of enzymes but also a preferential effect on lipolytic ones and such a complex process seems unlikely. The most probable explanation is that the inhibition occurs at the sites of absorption and is due to a physical barrier lining the gut wall and thus diminishing absorption (Katoch et al. 1971). The preferential effect on fat may be due to the size of the particles in which form it is absorbed. Other digestion products, monosaccharides and amino acids, are of relatively small particle size and thus may pass through a barrier with greater ease than the micelles in which form fat is absorbed.

EXPERIMENT 3.2.

Objective

A TME determination was carried out in order to confirm the depression in AME_N noted in Experiment 3.1. Nitrogen retention and fat digestibility were again determined.

Experimental

Design

The experimental design was that of a randomised block with five replicates of nine dietary treatments which were as follows:-

1. Basal diet (Control)
2. 2.5g guar gum/kg basal diet
3. 5.0g " "
4. 7.5g " "
5. 10.0g " "
6. 12.5g " "
7. 15.0g " "
8. 17.5g " "
9. Starved control birds

Birds and Management

The procedure followed was that of the rapid bioassay developed by Sibbald (1976), with a modification to the original method described in Experiment 2.1.

To ensure complete removal of food from the gut, both the starvation period and the collection period were extended to 48 hours. Forty-five adult White Leghorn cockerels were individually caged and starved for 48 hours, prior to being tube-fed 25 g of an experimental diet according to a previously randomised plan. Five birds remained starved, forming the starved control group of birds maintained for the collection of endogenous losses. The birds were returned to clean cages with a clean tray placed underneath and the excreta voided in the 48 hours subsequent to feeding were quantitatively collected.

The excreta samples were freeze-dried, allowed to equilibrate to atmospheric moisture, weighed and analysed for gross energy, total nitrogen and fat content.

Diets

The diets used in this experiment were samples taken from those diets fed in Experiment 3.1.

Results

The results of the TME assay are presented in Table 3.3, together with the figures for apparent nitrogen retention and apparent fat digestibility. The TME values are corrected to zero nitrogen retention (TME_N) and are presented as MJ/kg dry matter. Apparent nitrogen retention and apparent fat digestibility are in terms of g/kg dry matter food intake. That nitrogen retention is negative is a feature of the experimental method, rather than the dietary treatments.

There are no significant differences in the TME_N values nor in the nitrogen retention or fat digestibility figures, as would be expected.

Discussion

Guar gum, it appears, severely depresses AME_N whilst not affecting the TME_N of a diet. The explanation of this somewhat illogical conclusion could well lie in the difference between the two assays. AME is known to vary with the level of feed intake, whereas during the TME assay the feed intake is constant. In Experiment 3.1, however, guar gum had little effect upon food intake. AME is proportional to 1/food intake but there was only a slight correlation between the two ($r = .2819$) which suggests that the difference in food intakes alone do not fully explain the severe depression in AME.

One major difference between the TME and AME assays is the time factor involved. The AME assay takes place over at least a three day period and generally occurs after a ten- to fourteen-day adaptation period. The TME assay involves a small input of diet on one occasion

Table 3.3. Mean TME_N, Nitrogen retention and Fat digestibility of diets containing Guar gum

Diet	1	2	3	4	5	6	7	8			
Guar gum g/kg	0	2.5	5.0	7.5	10.0	12.5	15.0	17.5			
TME_N MJ/kgDM	16.37	16.46	16.59	16.67	15.91	16.61	16.16	16.81	±SE	.28	NS
Nitrogen retention g/kg food intake DM	-58.59	-67.28	-60.00	-68.39	-56.56	-71.79	-64.36	-64.53	±SE	10.75	NS
Fat digestibility g/kg food intake DM	100.09	112.30	103.02	97.74	93.99	99.30	96.14	98.85	±SE	1.10	NS

NS = not significant

only, involving no previous exposure to the substance under experimentation. It is possible that the effects of guar gum are cumulative, becoming more severe over a period of exposure.

Another important difference between the two methods is the incorporation of EEL in the TME estimation. A standard value obtained from starved control birds is used in the estimation as it is generally accepted (Sibbald, 1976) that EEL is independent of the diet. However, if this were not so then the TME calculation could be influenced in such a way as to mask the true response in the energy balance.

EXPERIMENT 3.3.

Objective

This experiment was designed to determine whether or not the effect of guar gum upon chick growth and AME is cumulative, i.e. its severity being dependent upon the length of time of exposure to the guar gum.

Experimental

Design

Four treatments were randomly assigned to 44 birds, giving 11 replicates of each. The treatments were:-

1. Control diet for 3 weeks.
2. Control diet for 2 weeks, test diet for 1 week.
3. Control diet for 1 week, test diet for 2 weeks.
4. Test diet for 3 weeks.

Birds and management

Sixty day-old, PRC stock, male broiler chicks from the same hatch were wing-banded and randomly assigned to four treatments and reared in thermostatically controlled battery brooders. At 7 days of age, the diet of group 3 was changed from the control diet to the test diet and at 14 days of age the diet of group 2 was changed in the same manner. At 14 days of age the birds were moved into individual cages. Mortality, mainly during the first 7 days reduced the number of birds per group to 11.

An **AME assay** was carried out over a three day period from 18 to 21 days of age. Feed intakes over the three days were recorded and excreta collected quantitatively. The excreta samples were oven-dried, equilibrated to atmospheric moisture, weighed and analysed for gross energy and total nitrogen. Food and water were available *ad lib*. throughout the 21 days.

Individual body weights were recorded at 1, 7, 14 and 21 days of age.

Diets

The control diet was of the same formula as in the previous experiments in this section (see Table 2.1). The test diet consisted of the control diet containing 20 g guar gum/kg. The diets were analysed for dry matter, gross energy and total nitrogen.

Results

Intakes over the AME assay period and AME values are presented in Table 3.4. The AME values are corrected to zero nitrogen retention and are presented in terms of kg dry matter AME, MJ/kgDM.

Food intakes were only significantly depressed for the birds in group 4, i.e. those birds which had been on the test diet for 3 weeks. This was most likely due to smaller body size of these birds (see Table 3.5).

The AME_N of the control diet, containing no guar gum (i.e. treatment 1) was significantly higher than the AME_N of the other three treatments but there were no statistical differences between the three guar gum treatment groups.

Mean body weights at 1, 7, 14 and 21 days of age are presented in Table 3.5. It is apparent that the mean body weight is significantly reduced within 7 days of a bird first receiving the guar gum diet. The results also suggest that the response to guar gum becomes more severe within 14 days. At 21 days of age the mean body weight of birds in group 2 was significantly less than that of the birds on the control diet (group 1) but was still significantly greater than the body weights of those birds in the other two treatment groups. Also it appears that after 14 days the depression of body weight does not become more severe, since at 21 days of age the mean body weights of birds in groups 3 and 4 are not significantly different.

Discussion

The difference between the AME and TME results noted in the previous experiments is not due, it appears, to an increasing

Table 3.4. Mean intakes and AME_N values of a 20g/kg Guar gum diet after four different periods of exposure.

Treatment	1	2	3	4
Period of exposure, weeks	0	1	2	3
Intakes over AME assay, g	170^a	141^a	137^a	117^b
±SE	8	7	7	11
AME_N MJ/kgDM	13.44^a	11.24^b	11.18^b	11.93^b
±SE	.11	.40	.40	.31

a.b. = within a horizontal line, those figures with the same superscript are not significantly different.

Table 3.5. Mean body weights of birds after different periods of exposure to Guar gum (g).

Treatment	1	2	3	4
Period of exposure, weeks	0	1	2	3
Wt. at 1 day	40[a]	39[a]	40[a]	39[a]
±SE	.6	1.5	1.2	.9
Wt. at 7 days	99[a]	101[a]	109[a]	87[b]
±SE	3.3	4.2	3.3	4.2
Wt. at 14 days	238[a]	251[a]	212[b]	184[b]
±SE	9.4	11.5	7.5	12.0
Wt. at 21 days	452[a]	376[c]	318[b]	288[b]
±SE	19.6	16.3	15.1	26.5

a.b.c. = within a horizontal line, those figures with the same superscript are not significantly different.

severity of the effects of guar gum over a period of time. The response to guar gum occurs rapidly, within one week, as illustrated both by the AME results and the mean body weights, and does not increase in severity on further exposure, at least in the case of AME values. The apparent increase in the severity of the effects of guar gum upon body weights may not have been a real effect. The mean body weight of the birds in group 2 at the onset of the guar gum treatment may have been sufficiently high to remain significantly greater than the mean weights of those birds on treatments 3 and 4 despite the depressing effects of guar gum. Figure 3.1 illustrates the rapid response to guar gum as seen by the decrease in rate of weight gain (represented by the slope of the lines) observed following the exposure to the test diet.

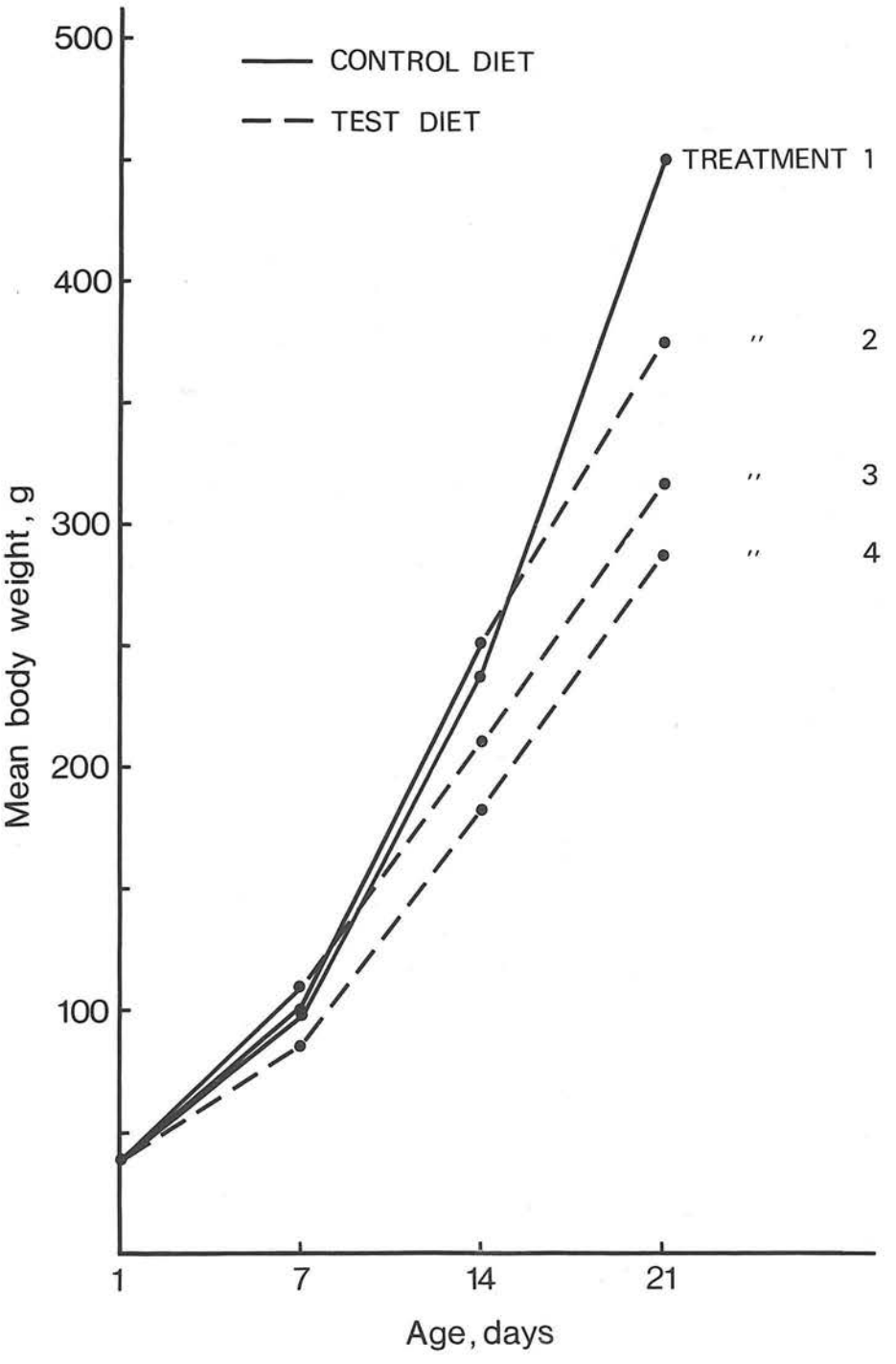

Fig. 3.1 Body weights of birds exposed to guar gum for 0 to 21 days.

EXPERIMENT 3.4.

Objective

A second determination of TME, using the relationship:-

TME = AME + EEL/Food intake

was carried out to confirm the lack of effect of guar gum on TME as indicated in Experiment 3.2.

The effect of guar gum upon EEL was also investigated. A possible explanation for the discrepancy between the effects of guar gum on AME and TME, as suggested by Experiments 3.1 and 3.2, is that guar gum influences EEL.

Experimental

Design

The design was that of a randomised block, involving five replicates of five dietary treatments. The treatments were:-

1. Basal diet (Control)
2. 5g guar gum/kg basal diet
3. 10g " "
4. 15g " "
5. 20g " "

Birds and management

About 150 day-old, Marshall stock, male broiler chicks from the same hatch were wing-banded and reared to 7 days of age in thermostatically controlled battery brooders. At 7 days of age they were individually weighed; 75 were selected from the middle weight band and distributed to 25 cages, 3 birds per cage, according to a previously randomised plan. The five dietary treatments were randomly assigned to the 25 cages, giving 5 replicates of each. Rearing continued for a further 10 days and this was followed by a three day AME assay. Intakes per cage over the three day period were recorded and all excreta quantitatively collected. Food and

water were available ad lib. at all times.

The birds were then starved for 48 hours, to ensure removal of food from the gut, and then starved for a further 24 hours and the excreta presumed to contain only material of endogenous origin were collected.

All the excreta samples were oven-dried, allowed to equilibrate to atmospheric moisture, weighed and analysed for gross energy and total nitrogen.

Diets

The basal diet (Control) was compounded to the same formula as in Experiment 3.1. The test diets consisted of the basal diet containing the appropriate concentration of guar gum.

The diets were presented in mash form and were analysed for dry matter, gross energy and total nitrogen.

Results

The AME was calculated from the equation:-

$$AME = \frac{(E_{in} - E_{out})}{\text{Food Intake}}$$

The TME was calculated from the relationship:-

$$TME = AME + \frac{EEL}{\text{Food Intake}}$$

The EEL value used was the gross energy of the excreta from the appropriate starved birds multiplied by a factor of 3 since the energy balance study took place over 3 days. All values were corrected to zero nitrogen retention. AME and TME are expressed in terms of kg dry matter (kg DM).

The results are presented in Table 3.6 and show a significant depression in AME_N, EEL_N and TME_N ($p<0.001$, 0.002, 0.01, respectively). There were no significant differences in the mean food intakes.

Discussion

Both AME_N and EEL_N were significantly depressed by the presence

Table 3.6. Mean food intakes, AME_N, EEL_N and TME_N values of diets containing guar gum

Diet		1	2	3	4	5	
Guar gum	g/kg	0	5.0	10.0	15.0	20.0	
Intake	g	188	190	179	177	171	NS
	±SE	2.3	5.8	4.1	1.5	4.1	
AME_N	MJ/kgDM	13.59	12.99	12.75	12.27	11.50	***
	±SE	.03	.08	.07	.29	.16	
EEL_N	kJ/72hrs	423	288	286	289	317	****
	±SE	45	20	8	12	21	
TME_N	MJ/kgDM	14.34	13.53	13.28	12.82	12.12	**
	±SE	.10	.10	.05	.28	.14	

NS = not significant
** = $p<0.01$
*** = $p<0.001$
**** = $p<0.002$

of guar gum, and the TME_N followed the same pattern of depression, although not to the same degree of significance. That the TME was reduced at a lower level of significance was probably due to a difference in food intakes which, although not statistically significant, would have affected the term EEL/Food intake. Or it may also be due to the effect on EEL "wearing off" because of the time gap between the feeding and measuring EEL.

The EEL is, therefore, depressed by guar gum and this is the most likely explanation of the discrepancy between the AME and TME noted in Experiment 3.2. Since the EEL is depressed by guar gum, during the TME assay the standard value of EEL obtained from the starved birds may be higher than the real EEL experienced by the birds. This would artificially raise the TME, possibly masking any real depression in the energy balance.

The EEL is generally considered to be independent of the diet (Sibbald, 1976). These results strongly disagree with this hypothesis and there would appear, therefore, to be a serious flaw in the use of the TME bioassay in the study of feedstuffs containing EEL depressing substances, such as guar gum.

DISCUSSION

Residual guar gum has been identified as the major contributor to the poor performance of chicks fed on guar meal (Kratzer + Vohra, 1963; Nagpal, 1968, Verma, 1977). The studies reported in this thesis were carried out to demonstrate the growth depressing action of guar gum and to more closely identify the cause of this action by examining the effect of guar gum upon the metabolisable energy values of diets, nitrogen retention and fat digestibility.

The inclusion of guar gum in diets for young broilers proportionately reduces mean body weight gain, dietary AME, nitrogen retention and apparent digestibility of fat. Feed intakes of the birds are not reduced by the presence of dietary guar gum, except possibly after a long period of exposure to high levels (e.g. 20g/kg for 21 days. Therefore, it can be concluded that the depression in body weight gain is likely to be a consequence of the reduction in dietary AME values.

Body weights were reduced to about 68% of the weights of control birds by 17.5 g guar gum/kg after exposure to the diets for fourteen days. Weight gains were reduced further, to 64% of those of control birds, by 20 g guar gum/kg over a twenty-one day period. This agrees well with other data (Vohra + Kratzer, 1964) which showed a depression in the growth of birds fed a diet contining 20 g/kg guar gum, to 61-67% of the weight of control birds. In these experiments, however, a growth depression was detected at the 2.5 g/kg level, in contrast to the studies reported in this thesis where a depression in growth was not detected until a dietary level of 7.5 g guar gum/kg was fed. White et al. (1978) demonstrated reduction in weight gains with 15 g/kg dietary guar gum, together with an increase in the occurrence of sticky droppings. Although no measurements were made during the studies presented in this thesis, sticky droppings were noted with birds receiving 10 g/kg guar gum, or more, increasing in severity as the dietary level of guar gum increased.

The lack of any significant effect on food intakes, except on a 21 day exposure to 20 g/kg guar gum is in agreement with the findings of Vohra et al., (1979). These authors reported a significant growth

depression ($p<0.01$) and a significant reduction in dry matter digestibility ($p<0.01$) when broiler chicks were fed on a diet containing 20 g guar gum/kg in place of maize starch; no effect upon feed intakes was observed. Vohra + Kratzer (1964) however, stated that the feed conversion efficiency of chicks fed a diet containing 10 g/kg guar gum was equivalent to that of control birds. Because the body weight gains of these birds were depressed, this would imply a parallel depression in food intakes. The experiment by these workers took place over a twenty-one day period but so too did the experiment of Kratzer et al. (1967) who reported no significant effect upon intakes of a diet containing 20 g guar gum/kg. Unfortunately, it is not possible to determine the level of significance of Vohra + Kratzer's results. Numerical reductions in feed intakes have been observed in experiments described in this thesis. This is not surprising in as much as food intakes are dependent upon body weight and body weights are reduced by the action of guar gum.

It has been concluded that the depression in body weight is a result of a reduction in the AME values of the diets containing guar gum. It can be further concluded that this reduction in AME is caused by a combination of the effects of guar gum upon both nitrogen retention and fat digestibility. Inclusion of 17.5 g guar gum/kg reduced nitrogen retention by 21% and fat digestibility by 22%. These observations support the conclusions made by Kratzer et al. (1967) who suggested that the main effect of guar gum was upon nitrogen retention because the greatest aggravation to growth occurred when guar gum was included in a high protein diet. This hypothesis is supported by reports that guar gum inhibits amino acid absorption in vitro (Katoch et al., 1971) and preferentially inhibits that of the sulphur-containing amino acids. If this phenomenon occurs in vivo it could lead to an imbalance in the net availability of amino acids for protein synthesis and thereby affect performance. However, the work in this thesis suggests that the main effect of guar gum is upon fat digestibility. This was concluded after statistical analysis of the data and involved the feeding of a conventional balanced diet only. Under different feeding circumstances the results may have differed. However, this does not detract from the original conclusions.

A further and most interesting feature of the AME studies is that the results indicate that guar gum has an ME value for chicks. Using the data from Experiment 3.1, linear regression (Figure 3.2) of all eight points (0-17.5 g/kg guar gum) gives the equation $y = 13.14 - 0.0636x$ ($r = -0.98$). This implies that the AME value for the control diet containing 0 guar gum/kg has an AME value of 13.14 MJ/kg, compared to the 12.98 MJ/kg as determined in the experiment. If only the data from the guar gum containing diets are used, then this difference becomes more pronounced; linear regression of 7 data points (2.5 - 17.5 g/kg) gives the equation $y = 13.25 - 0.0727x$ ($r = -0.99$), yielding an AME value for a diet containing no guar gum of 13.25 MJ/kg. These figures suggest that guar gum has an ME value of 160 to 270 kJ/kg (see Figure 3.2). Although Booth et al. (1963) reported that 76% guar gum was digested by rats when fed at a level of 60 g/kg, there have been no reports of guar gum digestion by poultry.

Vohra + Kratzer (1964) found that the growth inhibiting effect of guar gum could be removed by hydrolysing the guar gum prior to feeding with either an enzyme isolated from sprouted guar beans or a commercial preparation, cellulase-100. There have, however, been no reports of cellulase activity in the digestive juices of poultry. It would have been of great interest to have carried out a TME determination on pure guar gum but this was thought to be too difficult in practical terms, not least in that guar gum gelates on contact with water and may have done this within the gut, causing blockages. This property would also have caused problems in the actual tube-feeding.

A possible alternative explanation for the results of the ME studies is not that guar gum has an ME value but that it is reducing the EEL from the birds rather than contributing to the available energy of the diet. A major assumption involved in the TME determination is that endogenous energy loss is independent of feedstuff or its level of intake (Sibbald, 1975, 1976). However, Tenesaca & Sell (1981) have reported an increase in EEL with the feeding of increasing amounts of indigestible silica gel and suggested that this was a consequence of accelerated secretion of digestive enzymes and juices and/or exfoliation of intestinal mucosa. These results were obtained both when silica gel was tube-fed on its own or in combination with maize. In contrast to these findings Sibbald has demonstrated that neither sand, cellulose (1980a) nor sawdust (1981) had any effect on EEL.

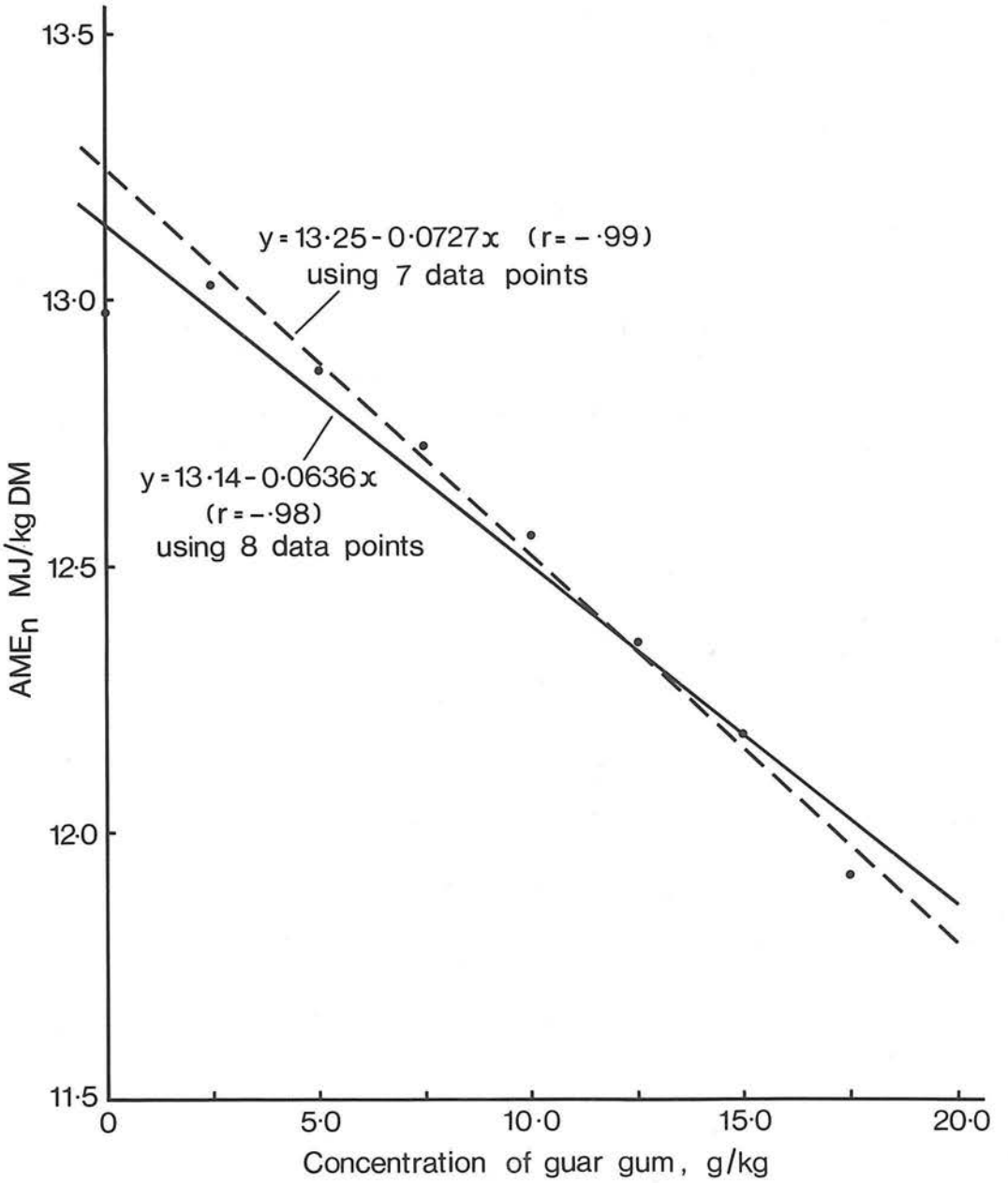

Fig. 3.2 Regression of AME_n data from Experiment 3.1 showing extrapolated values for diet containing 0 g guar gum g/kg.

Reports earlier in this thesis, involving both sand and cellulose, would appear to support Sibbald's hypothesis. To date there have been no reports of substances reducing EEL. The action of guar gum upon EEL may be, in fact, a reversal of the suggestion made by Tenesaca & Sell (1981) i.e. involving the decrease in secretion of digestive juices and/or exfoliation of intestinal mucosa, and may be intrinsically connected with the mechanism of action of guar gum.

It has been suggested (Katoch et al. 1971) that guar gum reduces absorption by creating a physical barrier between the nutrients in the feed and the absorptive surfaces of the gut. An increase in the viscosity of gut contents of rats fed diets containing guar gum, particularly of the fluid layer adjacent to the mucosa, has been demonstrated (Leeds et al. 1979; Johnson & Gee, 1980; Blackburn & Johnson, 1981). This would have various consequences. A physical barrier in the duodenum would reduce absorption of each of the major food groups. The final breakdown of protein to amino-acids occurs at the epithelial cells, tri- and dipeptidases being located in the brush border, as does the final breakdown of carbohydrates from disaccharides into monosaccharides. Micelles, the absorptive forms of fat, are also taken up at the brushborder. Thus, if guar gum creates a barrier to these absorptive processes, whole diet ME would be reduced. The suggested preferential effect of guar gum upon fat absorption could be explained at this stage as being a factor of particle size. Micelles have somewhat larger particle size than either disaccharides or peptides, and so may have more difficulty in passing through a physical barrier than the smaller molecules have. A physical barrier may also decrease defoliation of the mucosal surface, thereby reducing the EEL. The effect on EEL may also, however, be by a more complicated process.

In mammals, the presence of food, especially amino acids and fatty acids, in the upper small intestine causes the release of cholecystokinin-pancreozymin (CCK-PZ) and secretin from the mucosal cells. The presence of CCK-like peptides have been demonstrated in the avian gut (Dockray, 1979). CCK-PZ stimulates both the release of bile from the gall-bladder and pancreatic exocrine secretion (Bell et al. 1976). If a physical barrier inhibited the release of CCK-PZ this would not only reduce digestion due to the reduced availability of digestive enzymes but would also be expected to

reduce the bile and digestive juice fraction of the endogenous faecal component. Enteropeptidase, the main enzyme of succus entericus which activates the transformation of trypsinogen to trypsin, also originates from the microvillus membrane and its release, too, could be affected by a physical barrier. It is conceivable that the presence of guar gum creates a physical barrier to the absorptive surface of the gut, thereby not only reducing digestion as well as absorption but reducing the faecal components present in the endogenous energy excreted.

Studies reported in this thesis do not provide any direct evidence to support this hypothesis. Some pathological studies were carried out on birds which had received a diet containing 20 g guar gum/kg for twenty-one days, but no gross abnormalities of any organs were noted. Examination of some histological sections of intestine hinted at the presence of gum on the mucosal surface but, as the technique used also resulted in some fluorescence of the mucosa itself, the results were inconclusive (P.A.L. Wight, 1982, personal communication).

Future research should concentrate on establishing whether or not a physical barrier of gum lining the gut wall does occur in poultry; the work of Leeds et al. (1979), Johnson & Gee (1980), and Blackburn & Johnson (1981) was carried out with rats. The effect of guar gum upon intestinal secretions should be investigated, especially those of the gall bladder and pancreas because these contain enzymes responsible for the digestion of all food groups, but the gall bladder is especially involved in the digestion and absorption of fat. It was concluded earlier that a growth depressing response was observed with as little as 7.5 g guar gum/kg. If guar meal contains approximately 140 g guar gum/kg (c.f. literature review) then less than 55 g guar meal/kg diet could induce adverse effects. Various workers have shown that the effect of guar gum on performance can be reduced by both enzyme supplementation (Anderson & Warnick, 1964; Vohra & Kratzer, 1965; Verma, 1977) and physical treatments (Borcher & Ackerson, 1950; Vohra & Kratzer, 1964a). To encourage more extensive use of guar meal these processes must be economical to carry out on a large scale. An ideal solution to the problem caused by the presence of dietary guar gum would be for the gum industries to improve the efficiency of its extraction from the guar bean, thereby improving their own profits and producing a by-product more suitable for use as a feedstuff for poultry.

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5. APPENDIX

Methods of Analysis

The Determination of Gross Energy

Gross energy determinations of both diets and faeces were carried out using a PARR 1241 Automatic, Adiabatic Calorimeter, in conjunction with a Parr Calorimeter Master Control. Generally, approximately 1 g samples were formed into pellets, but in cases where faecal samples would not easily pellet the following technique was employed.

A gelatine capsule (EMscope Laboratories Ltd) was weighed and filled with the sample of faeces (approx. 1 g) and reweighed. The normal procedure was then followed. The G.E. of the faecal sample was then determined by the subtraction of the heat of combustion of the gelatine capsule from the total heat of combustion of the combined capsule and faeces, viz.

G.E. faeces = [(Energy equivalent of bomb x temperature rise) - G.E. capsule x wt capsule)] /wt of faeces

The G.E. of the gelatine capsules was obtained by a determination on a number of capsules compressed into one capsule.

Total Nitrogen

Total nitrogen of both diets and faeces was determined by the method described by The Fertilisers and Feeding Stuffs (Amendment) Regulations, 1976, for the Determination of Protein. For faecal samples however a sample weight of 0.2 - 0.3 g was used.

The Determination of Fat

The fat content of diets was determined by the method described by The Fertilisers and Feeding Stuffs (Amendment) Regulations, 1976, for the Determination of Oil - in the absence of milk powder.

The fat content of faeces was determined by the following procedure.

Acid Ether Extraction Method For Determining Fat in Faeces

In faecal samples a considerable quantity of fatty acids are present as soaps of various elements, principally calcium. These

soaps are not extracted by petroleum ether alone, but if the sample is acidified, the free fatty acids in the soaps are released and become extractable by petroleum ether.

The acidification and extraction is readily achieved by using a mixture of glacial acetic acid and petroleum ether 40/60, containing 10% by volume of acetic acid.

The sample of dry powdered faeces is weighed into an extraction thimble in the usual manner and placed in a soxhlet extractor. Fill the soxhlet extractor with the acetic acid - petroleum ether mixture and allow the liquid to syphon into the flask and then refill. Allow to stand for 12 hours (or overnight). Switch on the heating apparatus and extract the faeces in the usual way for 24 hours.

After extraction is complete remove as much solvent as possible from the flask by distilling into the empty soxhlet and finally dry on a rotary evaporator at 40°C. Dry for 10-15 minutes after the flask appears to be dry to ensure removal of all the acetic acid. To complete the process dry in an oven at 105°C. Cool in a desiccator and weigh.

Determination of Dry Matter

The dry matter content of diets was determined in the following manner. Duplicate 5 g samples were weighed into previously weighed containers with air-tight lids. The uncovered container and lid was then placed in an oven at 105°C and left overnight. The lids were replaced on the containers which were then cooled in a desiccator and weighed. The dry matter is the new sample weight expressed as a percentage of the original weight.

Tables of Compositions

PRC Vitamin Supplement No.3

Vitamin A	120 million i.u.
Vitamin D3	16 " i.u.
Vitamin E	500 g
Menaphthone	26 g
Riboflavin	80 g
Nicotinic acid	560 g
Pantothenic acid	200 g
Anti-oxidant	as necessary
Ground maize to make	50 kg

PRC Vitamin Supplement No. 4. (Chick grower, broiler)

Vitamin A	40 million i.u.
Vitamin D	12 " i.u.
Vitamin E	500 g
Menaphthone	26 g
Riboflavin	80 g
Nicotinic acid	560 g
Pantothenic acid	200 g
d - Biotin	1 g
Anti-oxidant	as necessary
Ground maize to make	50 kg

PRC General purpose Mineral Supplement No.5

Cu (as cupric sulphate)	70 g (fine crystals)
I (as potassium iodate)	8 g
Fe (as ferrous sulphate)	1600 g
Mg (as carbonate)	6000 g
Mn (as carbonate)	2000 g
Zn (as oxide)	1000 g
Ground maize to make	50 kg

Gross energy, total nitrogen and dry matters of diets used in Experiments 2.1 and 2.2

Diet	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Dilution g/kg	0	20	40	60	20	40	60	20	40	60	20	40	60	20	40	60
Dilutent	sand				cellulose			kaolin			sawdust			CKD		
G.E. MJ/kg DM	20.03	19.54	19.21	18.74	19.84	19.80	19.94	19.20	19.04	18.07	19.65	19.78	20.04	18.93	19.47	18.61
Total nitrogen g/kg	34.78	34.94	34.13	33.42	33.31	33.48	33.11	35.00	34.20	32.41	33.65	34.69	33.42	35.20	34.40	33.97
Dry matter g/kg	878.9	894.6	895.7	896.3	886.2	891.0	890.7	899.2	898.1	903.7	889.8	894.6	889.9	885.3	893.5	902.2

Gross energy, total nitrogen and dry matters of diets used in Experiment 2.5

Diet	1	2	3	4	5
	control	cellulose		sand	
Dilution g/kg	0	40	60	40	60
G.E. MJ/kg DM	15.27	15.07	15.25	14.27	16.66
Total nitrogen g/kg	33.2	31.50	31.80	30.70	30.90
Dry matter g/kg	896.50	897.7	898.20	898.80	898.50

Gross energy, total nitrogen and dry matter of diets used in Experiment 2.7

Starter diets

Diet	1	2	3	4	5	6	7	8
	control		sand-diluted			ME CP reduced		
GE MJ/kg DM	19.72		19.33	18.89	18.45	19.75	19.77	19.38
Total nitrogen g/kg	34.40		33.30	32.60	32.50	32.60	32.30	31.70
Dry matter g/kg	895.90		896.70	900.40	899.30	892.80	891.90	893.40

Finisher diets

Diet	1A	2A	3A	4A	5A	6A	7A	8A
	control		sand-diluted			ME CP reduced		
GE MJ/kg DM	18.78		18.90	18.23	17.59	18.90	19.03	18.80
Total nitrogen g/kg	30.40		31.20	30.10	29.10	30.20	29.10	28.50
Dry matter g/kg	878.10		879.20	884.40	889.40	875.30	875.30	878.00

Gross energy, total nitrogen, fat content and dry matter of diets used in Experiments 3.1 and 3.2

Diet	1	2	3	4	5	6	7	8
Guar gum g/kg	0	2.5	5.0	7.5	10.0	12.5	15.0	17.5
G.E. MJ/kg DM	19.54	19.47	19.68	19.69	19.62	19.63	19.58	19.69
Total nitrogen g/kg	37.60	37.00	37.80	37.40	37.10	38.10	37.40	36.30
Fat g/kg	105.00	115.00	107.00	105.00	102.00	104.00	104.00	104.00
Dry matter g/kg	927.00	924.60	921.00	921.70	926.40	924.40	926.40	923.70

Gross energy, total nitrogen and dry matter of diets used in Experiment 3.3

Diet	1	2
	control	test
Guar gum g/kg	0	20.0
GE MJ/kg DM	19.22	19.12
Total nitrogen g/kg	37.50	36.40
Dry matter g/kg	912.50	917.70

Gross energy, total nitrogen and dry matter of diets used in Experiment 3.4

Diet	1	2	3	4	5
Guar gum g/kg	0	5.0	10.0	15.0	20.0
GE MJ/kg DM	19.34	19.35	19.36	19.35	19.39
Total nitrogen g/kg	37.10	38.20	36.70	36.10	36.60
Dry matter g/kg	921.20	917.8	916.80	921.00	917.00

Tables of Analysis of Variance

In the following analysis of variance (ANOVA) tables

df	=	degrees of freedom
F	=	variance ratio
MS	=	mean square
NS	=	not significant
*	=	significant at P 0.05
**	=	significant at P 0.01
***	=	significant at P 0.001
SS	=	sum of squares

Analysis of Variance tables, Experiment 2.1

Source of variation	df	SS	MS	F	
Weight gains					
Blocks	2	2968.0	1484.0	3.45	*
Diluents	4	6260.9	1565.2	3.64	**
Levels	2	3381.8	1690.0	3.93	*
Diluent x Level	8	2861.0	357.6	0.83	
Control vs. rest	1	4757.8	4757.8	11.06	**
Residual	30	12908.9	430.3		
Total	47	33138.5			
Basal food intake					
Blocks	2	3126.3	1563.2	1.76	
Diluents	4	2361.0	590.2	0.67	
Levels	2	2390.8	3695.4	4.16	*
Diluent x Level	8	7314.6	914.3	1.03	
Control vs. rest	1	5476.8	5476.8	6.17	*
Residual	30	26628.8	887.6		
Total	47	52298.2			
Fce					
Blocks	2	.00625	.00312	5.14	*
Diluents	4	.00874	.00218	3.60	*
Levels	2	.00152	.00076	1.25	
Diluent x Level	8	.00584	.00073	1.20	
Control vs. rest	1	.00146	.00146	2.41	
Residual	30	.0182	.00061		
Total	47	.042			

Analysis of Variance tables, Experiment 2.1 cont.

Source of variation	df	SS	MS	F	
FI over AME study					
Blocks	2	0.0368	0.0184	2.47	
Diluents	4	0.1826	0.0456	6.08	**
Levels	2	0.0442	0.0221	2.95	
Diluent x Level	8	0.0606	0.0076	1.01	
Control vs. rest	1	0.0085	0.0085	1.13	
Residual	30	0.2240	0.0075		
Total	47	0.5566			
Ein - Eout					
Blocks	2	0.1247	0.0824	2.94	
Diluents	4	0.5776	0.1444	6.82	***
Levels	2	0.1049	0.0524	2.47	
Diluent x Level	8	0.2386	0.0298	1.41	
Control vs. rest	1	0.0289	0.0289	1.36	
Residual	30	0.6356	0.0212		
Total	47	1.7103			
AME					
Blocks	2	0.0093	0.0046	3.31	
Diluents	4	0.0265	0.0066	4.71	**
Levels	2	0.0066	0.0033	2.36	
Diluent x Level	8	0.0269	0.0034	2.43	*
Control vs. rest	1	0.0018	0.0018	1.31	
Residual	30	0.0420	0.0014		
Total	47	0.1130			

Analysis of Variance table for $\hat{TME}$, Experiment 2.1

Source of variation	df	SS	MS	F	
Different intercept for each treatment	15	.01802	.00120	2.42	*
Regression on food inputs:					
Single slope	1	.72198	.72198	1455.6	***
Diluents	4	.00522	.00131	2.64	
Levels	2	.00441	.00221	4.46	*
Diluent x Level	8	.00939	.00117	2.36	
Control vs. rest	1	.00157	.00157	3.17	
Residual	16	.007934	.000496		
Total	47	1.7103			

ANOVA Table. Experiment 2.2. - TME of Sand- and Cellulose-diluted diets

	Sand-diluted diets			Cellulose-diluted diets		
Source of variation	df	MS	F	df	MS	F
Dilution	3	0.304639	0.871 NS	3	0.258867	1.928 NS
Residual	19	0.349639		15	0.134299	
Total	22			18		

ANOVA Table. Experiment 2.3. - Effect of Sand upon EEL

Source of variation	df	EEL MS	EEL F	EEL_N MS	EEL_N F
Treatment	3	241.764	0.950 NS	99.3806	0.829 NS
Residual	20	254.458		119.885	
Total	23				

ANOVA Table. Experiment 2.4. - Length of Caeca

Source of variation	df	Right Caeca MS	F	Left Caeca MS	F
Treatment	2	0.926334	0.566 NS	0.376333	0.256 NS
Residual	27	1.63559		1.47267	
Total	29				

ANOVA Table. Experiment 2.6. - Effect of Cellulose upon $EEL_{(N)}$

		INTACT				CAECECTOMISED			
		EEL		EEL_N		EEL		EEL_N	
Source of variation	df	MS	F	MS	F	MS	F	MS	F
Treatment	2	586.332	2.749 NS	15.5037	0.132 NS	1775.44	5.159 *	543.111	8.442 *
Residual	8	213.282		117.557		344.121		64.338	
Total	10								

Analysis of Variance Table. Experiment 2.7

Source of Variation	df	log-food intake at 4 weeks MS	F	log-food intake at 8 weeks MS	F
Between Blocks					
Sex	1	0.0841953	85.537 ***	0.2015645	48.350 **
Residual	4	0.0009843	1.472	0.0041688	8.848
Total	5	0.0176265	26.355	0.0436480	92.641
Within Blocks					
Dilution	3	0.0022397	3.349	0.0043659	9.266
Diet	1	0.0253067	37.838 **	0.193233	41.013 **
Sex-dilution	3	0.0010011	1.497	0.0004764	1.011
Sex-diet	1	0.0006597	0.986	0.0005833	1.238
Dilution-diet	3	0.0045825	6.852 **	0.0085682	18.185 ***
Sex-dilution-diet	3	0.0019757	2.954	0.0004007	0.850
Residual	28	0.0006688		0.0004712	
Total	42	0.0017641		0.0017746	
Grand Total	47				

Analysis of Variance Table. Experiment 2.7

Source of Variation	df	log-Body weight at 4 weeks MS	F	log-Body weight at 8 weeks MS	F
Between Blocks					
Sex	1	0.0664728	25.292 **	0.3277892	54.465 **
Residual	4	0.0026282	3.519	0.0060183	7.624
Total	5	0.0153971	20.619	0.0703725	89.144
Within Blocks					
Dilution	3	0.0010436	1.398	0.0011566	1.465
Diet	1	0.0055987	7.497	0.0145240	18.398
Sex-dilution	3	0.0016651	2.230	0.0008245	1.044
Sex-diet	1	0.0000826	0.111	0.0002343	0.297
Dilution-diet	3	0.0031625	4.235 *	0.0040677	5.153 **
Sex-dilution-diet	3	0.0012892	1.726	0.0001517	0.192
Residual	28	0.0007467		0.0007894	
Total	42	0.0011446		0.0013206	
Grand Total	47				